Dragonflies as bioindicators and biodiversity surrogates for freshwater ecosystems

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Declaration

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General summary

Biological indicators (bioindicators) are useful for rapid and cost-effective ecosystem assessments. Dragonflies are valued for their potential as bioindicators in freshwater ecosystems. My dissertation aims to assess and expand on their use as bioindicators in transformed landscapes and as surrogates for other aquatic biodiversity. Of the three bioindicator categories (environmental, ecological and biodiversity), biodiversity indicators and their application are poorly understood. The umbrella species concept is a biodiversity surrogacy method that aims to conserve a large number of species in an ecosystem by focusing on a select group of co-occurring species. I used the umbrella index, which quantitatively identified a group of seven dragonfly species and a group of eight Ephemeroptera, Plecoptera and Trichoptera (EPT) species, any of which could be used as biodiversity surrogates (Chapter 2).

Adult dragonflies can only be surveyed on warm, windless days during summer, but are easily identifiable. On the other hand, their larvae can be sampled under any weather conditions and are also sensitive bioindicators. I showed that the interchangeability of the life stages for assessments was dependent on landscape spatial scale, coupled with the specific question asked (Chapter 3). Comprehensive biodiversity surveys at fine ecological scales should sample both adults and larvae. However, at larger spatial scales with coarser ecological questions, either adults or larvae can be used.

To mitigate the detrimental effects caused by forestry, ecological networks (ENs) are integrated into plantation landscapes. These comprise grassland corridors connected to protected areas (PAs), which often include rivers. They aim to conserve biodiversity by creating habitats or facilitating dispersal of grassland species. I showed that water quality and adult dragonfly diversity did not differ between EN corridors and PAs (Chapter 4). Therefore, the EN approach is an effective method for conserving dragonfly diversity and river ecosystem integrity in plantation landscapes.

In the Pietermaritzburg Botanical Gardens, an insect conservation pond was built along a degraded stream. Dragonfly species richness and abundance significantly increased, as both lentic and lotic species were able to colonize the area. Over time, the pond became overgrown and siltation reverted it back to a stream, which negatively affected dragonfly diversity. Shortly after extensive restoration efforts, the dragonfly assemblage had almost completely recovered

and closely resembled that of the original pond. This was linked to alien plant removal, decreased vegetation cover and the inclusion of a range of microhabitats. This indicates that conservation ponds need to be actively maintained to keep their function as biodiversity reservoirs. This highlights the value of dragonflies as indicators of habitat quality in aquatic restoration projects.

Throughout the thesis, adult dragonflies continuously demonstrated their success as bioindicators. The umbrella index validated the use of dragonflies as biodiversity indicators and surrogates for some of the most sensitive aquatic taxa (the EPT). Although dragonfly larvae and adults are both indicators of water quality, they are not always interchangeable, in terms of sampling at the species level. Their interchangeability depends on the specific question asked and the scale used. I also successfully monitored dragonfly assemblage responses in agricultural lotic and urban lentic environments, which highlighted their benefits for good aquatic conservation planning in transformed landscapes.

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Chapter 1: General introduction

1.1 Global threats to freshwater ecosystems

Freshwater covers only ~0.8% of the Earth's surface but is considered our most vital natural resource (Vörösmarty et al., 2010). Despite the importance of freshwater ecosystems, humans have degraded them to the point where they are now among the most threatened ecosystems on the planet (Vörösmarty et al., 2010; Boon & Raven, 2012). Approximately 10% of all described species inhabit freshwaters, with aquatic insects constituting a major proportion of the animal abundance (Gerten et al., 2008; Dijkstra et al., 2014; Harrison et al., 2016). However, the majority of research and public attention falls on vertebrates, and invertebrates are underrepresented (Ceballos et al., 2010; 2017). The severe decline of aquatic insects has put at risk the ecosystem goods and services they provide us with (Russi et al., 2013; Green et al., 2015). Some of these include herbivory, water filtration, decomposition of organic matter and as predators controlling pest species (Macadam & Stockan, 2015). Despite these, 33% are threatened with extinction, which is 5% higher than for terrestrial insects (Sánchez-Bayo & Wyckhuys, 2019).

Due to our dependence on water, human settlements are concentrated around freshwater bodies (Strayer, 2006). Impacts related to urban and agricultural development, are therefore the main threats to aquatic biodiversity (Wasson, 1996; Dudgeon et al., 2006; Vörösmarty et al., 2010; Habel et al., 2019). In particular, freshwater insect declines are strongly linked to activities that cause habitat loss, fragmentation or degradation (Revenga et al., 2005). Freshwater bodies, especially rivers, are affected by the terrestrial landscapes that surround them. Therefore, some of the worst effects result from activities that take place within the outer drainage basin, such as land clearing, livestock farming, vegetation removal, crop fertilization, soil erosion and building construction. Other effects have a more direct influence, such as dam construction, water extraction, fishing, alien invasive species introductions, pollutants and effluent inputs (Darwall et al., 2009; Harrison et al., 2018). Furthermore, the growing challenges associated with climate change cannot be ignored, as it has become a prominent threat to freshwater biodiversity (Dudgeon et al., 2006). Aquatic insects are extremely reliant on water availability and temperature, both of which are climate dependent. Events such as droughts can decrease water availability and results in habitat fragmentation, which can inhibit aquatic insect

dispersal. This is problematic when ecosystems become uninhabitable, due to climate-related changes, and insects need to escape to more suitable conditions (Woodward et al., 2010).

1.2 Threats to South African freshwater systems

Anthropogenic developments often require the removal of natural riparian vegetation, which provide important habitats and resources for biota. In urban areas, riparian vegetation is replaced with shallow-rooted, ornamental species or structures such as footpaths. In agricultural settings, riparian zones are ploughed and natural vegetation replaced with crops. Loss of riparian vegetation causes bank erosion, alters infiltration ability and increases sedimentation, pollutant inputs and runoff into rivers (Dallas & Day, 2007). Furthermore, opportunities for alien plant invasions are created (Bennett et al., 2001). Rivers in particular, are transport vessels for seeds of invasive plants, with *Acacia*, *Hakea*, *Pinus* and *Eucalyptus* tree species some of the most problematic plants in South Africa (Richardson, 1998; Richardson & van Wilgen, 2004). These invasive species spread quickly, increase shade cover, homogenize habitats and significantly reduce stream flow as they consume large quantities of water (Le Maitre et al., 1996; Richardson & Van Wilgen, 2004).

Infiltration capacity is reduced when soil is compacted or surfaces are covered with impermeable materials such as tar (Trombulak & Frissell, 2000; Alberti et al., 2007). Overland flow is then drastically increased and combined with inputs from drainage systems, putting immense pressures on the rivers they enter. Destruction of habitats occur when channels are deepened or lined with concrete, to maximise their intake capacity to cope with the excess water (May et al., 1997; Alberti et al., 2007). Alternatively, some urban and agricultural activities require vast quantities of water and over extraction is a common problem. Furthermore, pollution from organic matter, pesticides, fertilizers, drug residues and saline drainage frequently end up in rivers (De Wit et al., 2002; UNEP, 2016). Decreased water quality reduces the survival of sensitive biota that are dependent on these ecosystems (Nõges et al., 2016).

The Cape Floristic Region (CFR), in the Western Cape Province of South Africa, is a global biodiversity hotspot (Day & Day, 2009). A conflict of interest exists because the unique climatic and edaphic conditions responsible for the rich biodiversity of the region, are also optimal conditions for vineyard cultivation. Approximately 90% of South Africa's vineyards are farmed in this region (Fairbanks et al., 2004). The CFR is one of the six recognized floral

kingdoms, with 69% of its 9000 plant species endemic to the region (Goldblatt & Manning, 1999). Some farmers do not practice sustainable methods, and plough right up to the edge of rivers. The heavy machinery used to level river-beds destroys the natural riparian vegetation, leaving river banks bare. This increases erosion, and runoff of pesticides and fertilizers, which often end up in nearby rivers (Dabrowski et al., 2002). Furthermore, farming processes require significant quantities of water, which is problematic for a drought-stricken area.

The effects related to agriculture, depend on the crop type and intensity of farming. For example, the KwaZulu-Natal Province of South Africa, is a worldwide leader in the timber sector and has over 38.5% of its land allocated to plantations (Hurley et al., 2012). The area falls within the grassland biome of the Maputaland-Pondoland-Albany hotspot. This is one of the largest, but also most degraded biomes in the world (Neke & Du Plessis, 2004). The biggest concern is that the sites most suitable for plantations overlap with those that contain the highest natural grassland biodiversity (Lipsey & Hockey, 2010). The large-scale compositional and structural transformation from open, biodiverse grasslands to closed-canopy, exotic timber plantations cause severe habitat loss and fragmentation for biota (Murphy & Lovett-Doust, 2004; Neke & Du Plessis, 2004). Plantations comprise of *Pinus*, *Eucalyptus* and *Acacia* tree species (Edwards & Roberts, 2006). The shading effect from these alien trees inhibit growth of any other understorey vegetation. Other concerning effects associated with these exotic plantations, such as stream flow reduction, date back as far as 1915 (Dye & Versfeld, 2007).

1.3 Conserving freshwater ecosystems

Protected areas (PAs) are set aside to conserve the biodiversity of certain fauna and flora. Statutory PAs in South Africa include the Department of Forestry and Fisheries (DAFF) forest nature reserves, provincial reserves and national parks. These conservation areas are formally protected by national and provincial legislation (Frazee et al., 2003; Rouget, Richardson & Cowling, 2003). Non-statutory reserves comprise local and private nature reserves, conservancies, mountain catchment areas, DAFF demarcated forests and natural heritage sites. These areas have less stringent legal obligations and institutional support (Frazee et al., 2003; Rouget et al., 2003).

Less than 15% of the major rivers in South Africa are reasonably to well-represented in PAs (Nel et al., 2009). In particular, rivers are often incidentally conserved when terrestrial PAs are demarcated (Saunders et al., 2002). Management of PAs focus primarily on terrestrial

biodiversity, which can be to the detriment of aquatic ecosystems, for example when river-side lodges, roads and dams are constructed within PAs (Gaylard et al., 2003). Due to the flowing nature of rivers, their partial inclusion in PAs does not guarantee their conservation. Any negative influences outside the boundaries of PAs will impact river ecosystems (Mancini et al., 2005; Nel et al., 2009). Despite this, a national study showed that PAs have a greater proportion of intact river systems (50%) compared to rivers not in PAs (28%) (Nel et al., 2009). Rivers should therefore be incorporated into PA schemes in order to conserve them (Nel et al., 2009). The other half of the rivers currently included in PAs are of a poor quality because of upstream anthropogenic activities that fall outside of the borders (Nel et al., 2009). For this reason, to effectively conserve river ecosystems, management must extend beyond the boundaries of PAs and into transformed landscapes.

In KwaZulu-Natal, forestry companies aim to maintain structural, compositional and functional biodiversity by implementing ecological networks (ENs) (Samways et al., 2010; Pryke & Samways, 2015). These comprise areas of untransformed land with interconnected nodes and corridors, which ensure connectivity between natural habitat patches and even to PAs (Joubert & Samways, 2014; Samways & Pryke, 2016). Forestry industries have already allocated more than a third of their plantation holdings to establishing ENs. Many EN corridors include streams and rivers, which can be used for dispersal, resource provision or can function as habitats. Their role depends on the organism, as well as the size and condition of the corridor. Encouraging research has reported the effectiveness of ENs for conserving terrestrial arthropod diversity as well as do PAs (Pryke & Samways 2012a). They have effectively aided in the dispersal of organisms, including small mammals, butterflies (Haddad, 1999; Pryke & Samways, 2001) and bird-dispersed plants (Haddad et al., 2003). They have successfully maintained the diversity of many arthropod taxa, provided they are large enough to overcome edge effects (Pryke & Samways, 2012b; Samways & Pryke, 2016) and integrate natural landscape heterogeneity (Pryke & Samways, 2015). Well-designed ENs have also shown to support high dragonfly diversity (Kietzka et al., 2015; Pryke et al., 2015).

In cities, green spaces include open areas of human-modified or natural land with no urban infrastructure, often vegetated and encompassing natural features (Taylor & Hochuli, 2017). Ecologically-designed water infrastructures, like ponds, have both social and environmental benefits (Wendel et al., 2011). Some ponds are created purely for their aesthetic value (Gledhill et al., 2005), while others can provide important ecosystem services such as pollution

extraction or storm water control (Mallin et al., 2002; Hassall & Anderson, 2015). Conservation ponds can contribute significantly to biodiversity at a regional level (Hassall, 2014). For aquatic species that struggle to survive in the urban environment, ponds may act as stepping-stones enabling movement through the landscape to more favourable habitats (Simaika et al., 2016). More hardy species are able to survive in these ponds, and other synanthropic species actually benefit from the surrounding urban environment (Hassall, 2014). To maximize the efficacy of conservation ponds, knowledge of the focal taxa is crucial. For example, ponds have been constructed specifically for dragonfly conservation in South Africa (Steytler & Samways, 1995; Suh & Samways, 2001), Europe (Harabiš & Dolný, 2012; Maynou et al., 2017) and Japan (Primack et al., 2000). Vegetation-related characteristics significantly influence dragonfly diversity and are therefore important factors to consider in the design of such ponds (Steytler & Samways, 1995; Samways & Sharrat, 2010). Thereafter, pond management is crucial. Mismanagement and neglect will reduce biodiversity as habitat conditions deteriorate. For an aquatic insect conservation pond in South Africa, Suh & Samways (2005) based management recommendations on dragonfly habitat requirements. These included alien plant removal, rotational trimming of marginal vegetation, occasional pond dredging and limiting shade cover.

1.4 Assessing and monitoring with biological indicators

Biological indicators are species or groups of species that can reflect the biotic or abiotic condition of an ecosystem, can help detect the impact of environmental change on a community/ecosystem or are indicative of the diversity of certain taxa/habitats (Cairns et al., 1993; McGeoch & Chown, 1998; Gerhardt, 2002). They have been broadly divided into three categories: **Environmental indicators** can include physical parameters, such as salinity levels, aswell as biotic parameters, such as species richness. These have recognized responses to changes in environmental state or to environmental disturbances and signal early warnings of potential problems (Spellerberg, 1991; Bockstaller & Girardin, 2003). **Ecological indicators** are sensitive to the effects that specific environmental stresses have on the biotic components of ecosystems (Gerhardt, 2002). They are not simply gauges that reflect environmental changes, their response to a disturbance is representative of the responses of some of the other species that inhabit the same area (Jackson et al., 2000; Dale & Beyeler, 2001). In other words, they reflect responses between species. An important difference between environmental and ecological indicators is that for environmental indication the abiotic variable of interest is

usually something is easily measured, such as salinity or pH. Whereas, for ecological indication the state of interest is not as easily quantified, often due to it being more complex, such as with climate change or habitat degradation (McGeoch, 1998). Therefore, the variables to be indicated will have different degrees of susceptibility to measurement error and differing statistical properties, which will affect the performance of the bioindicator. **Biodiversity indicators** can represent the diversity of other taxa that occur in the same ecosystem (McGeoch, 1998; Gerhardt, 2002; Duelli & Obrist, 2003). A diversity measure such as species richness of a selected biodiversity indicator taxon can mirror the species richness of other taxa (Noss, 1990; Prendergast & Eversham, 1997).

Aquatic insects are the most widely used indicators of freshwater ecosystems (Lenat & Barbour, 1994; Bulánková, 1997; Bonada et al., 2006). Some of the commonly used taxa are the larvae of the Ephemeroptera, Plecoptera and Trichoptera (EPT), which are often grouped together and used as a biotic index in the northern hemisphere, as well as the adults and larvae of Odonata (dragonflies and damselflies, here collectively referred to as dragonflies). These taxa are sensitive to changes in their habitats and make good environmental and ecological indicators of freshwater conditions (Bulánková, 1997; Baker & Sharp, 1998; Magoba & Samways, 2010; Samways & Simaika, 2016; Masese & Raburu, 2017). The aquatic stages of dragonflies and the EPT are important components of indices like the South African Scoring System (SASS). This rapid bioassessment tool makes use of the indicator potential of variously sensitive taxa to assess freshwater health (Chutter, 1995; Dickens & Graham, 2002; Dallas 2004; 2013). Similarly, the Dragonfly Biotic Index (DBI) uses the presence of adult dragonfly species at a site to measure freshwater habitat conditions (Simaika & Samways, 2009; 2011). It has been recommended that larvae and exuviae be sampled when possible to improve the accuracy of the index (Samways & Simaika, 2016).

Adult dragonflies are highly mobile and can easily move from deteriorating, unfavourable habitat conditions to more suitable environments. Ecological integrity can thus be determined using attributes of the particular suite of dragonflies species present at a site. For the DBI, each species has a predetermined score based on the quantitative assessment of three sub-indices, ranging from 0 to 3 (Simaika & Samways, 2009; Samways & Simaika, 2016). These comprise the categorised geographical range of the species, its threat status, and its sensitivity to changes in habitat conditions (anthropogenic disturbance) (Simaika & Samways, 2009; 2011; Kietzka et al., 2015). The sum of these gives an overall score ranging from 0 to 9 for a given species

(Simaika & Samways, 2009; 2011; Samways & Simaika, 2016). Species with a score of 0 (0 + 0 + 0) would include those that are widespread, not threatened and highly tolerant of disturbance. Species with a score of 9 (3 + 3 + 3) would comprise those that are range restricted, threatened and extremely sensitive (Simaika & Samways, 2009; 2011, Samways & Simaika, 2016). The average DBI score at a site, total DBI score divided by the number of species present, can be used to compare freshwater conditions between sites (lentic or lotic) or to monitor change over time.

To date, the DBI has effectively assessed ecosystem integrity (Smith et al., 2007), prioritized sites for conservation action (Simaika & Samways, 2011; 2012; Harabiš & Dolný, 2012) and monitored the progress of stream restoration projects, after the removal of invasive alien trees (Samways & Taylor, 2004; Magoba & Samways, 2010). A strong correlation was found between SASS and DBI scores (Smith et al., 2007; Simaika & Samways, 2009; Samways & Simaika, 2016). Despite this, the DBI is the more sensitive index because it functions at the species rather than higher taxonomic level. Furthermore, SASS can only be applied to lotic water bodies, whereas the DBI can operate in both lentic and lotic conditions (Samways & Simaika, 2016).

Environmental indicators in freshwater ecosystems, and their application as indices, tell us about ecological health, which is often linked to water quality in freshwater ecosystems. However, water quality is a measure related to human consumption safety levels and does not indicate the state of biodiversity at a site (Graham, 1999). Therefore, by using environmental indicators alone to select sites for conservation action, there is no guarantee of conserving biodiversity. Biodiversity indicators represent the biodiversity of other taxa in the same ecosystem and should be incorporated into conservation planning. Regions and ecosystems with high biodiversity and where a large number of endemic species are present, should be prioritized, such as for aquatic insects in the CFR. The rivers of the region are a centre of endemism for dragonflies and some EPT species are so rare that they are restricted to single streams (Wishart & Day, 2002; Dallas & Day, 2007).

The umbrella species concept is a management-orientated, biodiversity surrogacy method that aims to conserve a number of co-occurring species in an ecosystem, by concentrating on a single or small group of species (Fleishman et al., 2000; Roberge & Angelstam, 2004; Bried et al., 2007). The exact origin of the concept is uncertain but Frankel and Soulé (1981) are said to be the first to use the term umbrella (Roberge & Angelstam, 2004). Carefully selected

umbrella species can identify biodiverse sites that require conservation action (Fleishmanet al., 2000; Bried et al., 2007). The umbrella index is a formula designed to quantitatively select suitable umbrella species (Fleishman et al., 2000; 2010). The calculated score for each potential umbrella species is based on its occurrence rate (number of sites it is recorded at), its co-occurrence with other species (number of sites where both occur) and its sensitivity to human disturbance (Fleishman et al., 2000; 2010). A good candidate umbrella species should ideally have 50% site occupancy, frequently occur with the focal taxa to be conserved and be more sensitive to anthropogenic disturbance than the majority of the other taxa (Fleishman et al., 2000; Bried et al., 2007). The index has successfully been applied to birds and butterflies (Betrus et al., 2005), and to dragonflies and wetland plants, in the USA (United States of America) (Bried et al., 2007). Thus far, dragonfly larvae have successfully predicted the diversity and structure of some aquatic macroinvertebrates assemblages (Briers & Biggs, 2003; Lee Foote & Rice Hornung, 2005) and are therefore suggested as umbrella taxa (Hornung & Rice, 2003; Schindler et al., 2003; Bried et al., 2007).

1.5 Using dragonflies as the focal taxon

A dragonfly's life cycle is characterised by an extreme habitat shift, from an aquatic insect to a terrestrial one (Corbet, 1999; Stoks & Cordoba-Aguilar, 2012). In order to define the value of a dragonfly species indicator potential it is crucial to understand its habitat requirements and distribution (Patten et al., 2015). The larvae, adults and the exuviae are all used in freshwater assessments and the most suitable stage to use depends on various factors such as the research questions, the geographical area of concern, sampling season and the degree of expert knowledge (Chovanec, 2000).

Although they have complex life cycles, with stages that inhabit very different environments, dragonfly larvae and adult stages are not necessarily independent (Sih et al., 2004; Bried et al., 2015a). Most adult male dragonflies are highly territorial of their waterside habitats (Moore, 1957; Corbet, 1980; Kemp, 2018). Mating and egg laying take place within these demarcated areas, which is why adult males and their conspecific larvae would be expected to occur within close proximity to one another (Buskirk & Sherman, 1985). Alternatively, species that breed in isolated habitats, like certain CFR rivers, may remain close to the sites from where they

emerged (Hawking & New, 1999; Angelibert & Giani, 2003; McCauley, 2007). This would strengthen the link between adult and larval habitat co-occurrence, and responses to the same environmental variables, as was the case for ponds in the UK (United Kingdom), where the majority of individuals remained at their natal ponds (Conrad et al., 1999). In addition to co-occurences, some studies have reported numerical carryover across metamorphosis, which may be evidence for exchangeability between the stages (Remsburg, 2011; Stoks & Córdoba-Aguilar, 2012). However, this does not stand for all studies regarding exchangeability of life stages but it is possible that some of these did not account for detectability differences when sampling the different stages (Hawking & New, 1999; 2002; D'Amico et al., 2004; Raebel et al., 2010; Giugliano et al., 2012; Bried et al., 2015a). Regardless of the life stage, it is important to consider the appropriate spatial scale to use for any general assessments using indicators (Boulton, 1999). Little is known in terms of the correct scale to use for dragonfly larvae. For the highly mobile adults, Samways and Simaika (2016) recommend sampling sites be 100 m in length but this would depend on the type of questions being investigated. Furthermore, whether the relationships between dragonfly larvae and adults are scale dependent is unknown.

Carryover effects between larvae and adults can also be ecological. Behavioural syndromes can carry over through metamorphosis from larvae through to adult. This has been shown regarding behaviour (for example active vs less active dragonflies) in individuals, which is a trade-off between increased encounters with predators and increased food intake (access to prey). Both adult boldness and activity correlated with the behaviour type of the larvae (Brodin, 2008). Dragonflies are also susceptible to ecophysiological carryovers. For example, damselfly larvae experienced decreased fitness when exposed to contaminants in their habitats, which carried over to the adult phase. These effects were exacerbated when individuals were faced with natural stressors such as extreme temperatures or predation (Bried et al., 2015b; Stoks et al., 2015). Alternatively, environmental variables could effect each life stage differently (Valente-Neto et al., 2016).

Adult dragonfly surveys are more popular than larval or exuvial surveys for water quality assessments. Adult, male individuals make ideal indicator candidates with their vibrantly coloured, conspicuous bodies making species-level identification easy (Simaika & Samways, 2009; Futahashi et al., 2015; Samways & Simaika, 2016). They are also easy to sample and do not require in-water netting like larval sampling does. Dragonfly surveys around farmland ponds in the UK showed that larval and exuvial surveys were interchangeable with each other

(Raebel et al., 2010). However, adults also utilize certain areas for feeding or could be just passing through rather than using them as breeding habitats (Simaika et al., 2016; Maynou et al., 2017). The use of adults in assessments has thus been criticised, as it may lead to unreliable estimates of reproductive populations (Hardersen, 2008; Raebel et al., 2010). Although they avoid positive bias, missed species can cause negatively biased species richness and false species absences (Bried et al., 2012).Residency can be assumed for adult Odonata species observed in mating habitat. This is because they generally copulate and oviposit only in viable freshwater habitats but only recording these individuals would be a severe underrepresentation. Exuviae and tenerals are sure indications of reproductive success and life cycle completion in an area (Raebel et al., 2010). However, tenerals are difficult to identify and to handle without damaging them and will disperse within a few hourse once their bodies have dried adequately (Patten et al., 2015). Exuviae, are inconspicuous and don't persist for a long period of time after emergence, especially in areas faced with adverse weather conditions (Lubertazzi et al., 2009). On the other hand, larvae are also challenging to sample from their in-water habitats where they are well camouflaged and can easily hide under rocks or in vegetation. Furthermore, a high level of expertise and knowledge of habitat preferences is vital for locating these life stages (Samways, 1992). They can also be difficult to identify to species level, which is not always possible, because in many countries the larvae of certain species are yet to be described. Individuals should ideally be in their final instar for accurate identification, which makes sampling season important. Aquatic sampling can also be unsafe if water conditions are turbulent or in areas where dangerous animals or diseases are present (Samways, 2008; Bird et al., 2019). Despite these challenges, including larvae and exuviae in studies on the ecology of dragonflies may provide additional data on habitat preferences that may be obscured by sampling only the adults (Samways & Simaika, 2016).

The actual number of resident species in an area lies somewhere between those recorded in high-biased adult observations and those recorded in low-biased larvae/exuviae surveys (Bried et al., 2015a). Bried et al. (2015a) defined criteria to deduce species residency, by relating adult dragonfly surveys to exuviae in the USA. Dragonfly species residency was accurately deduced when adult surveys met the provided criteria. Patten et al. (2019) used the presence of tenerals as indicators of breeding success and also related them to adult surveys. Their research aimed to determine thresholds for dragonflies in general (i.e. the minimum number of adults that would need to be observed in order to accurately confirm that a species is breeding in a certain area). No single threshold was found to be applicable to all dragonflies and varied

between suborders, families and genera. Although such methods may improve future freshwater applications, they are area and species specific and would need to defined accordingly and extensively tested.

1.6 Thesis aim and outline

In this dissertation, I aimed to determine the value of using dragonflies as bioindicators for assessing the responses of aquatic insect diversity to anthropogenic change. To achieve this, I first set out to explore whether adult dragonflies make successful biodiversity indicators for EPT larvae (and vice versa), tested in four CFR rivers (Chapter 2). For both dragonflies and EPT, I use the umbrella index to select two groups of potential surrogate species as representatives for the overall biodiversity of the groups. Given our current biodiversity crisis, there is an urgent need to test applications and search for shortcuts aimed at rapid conservation management. I expect that the umbrella index will perform adequately in selecting successful umbrella species and that the selected group of dragonflies will prove to be more successful as surrogate species than the selected EPT group for conveying the overall biodiversity of the groups. I think this success will be due to easier sampling of adult dragonflies and the application of species-level sensitivity knowledge.

I then determine the strength of the habitat associations and responses to environmental variables between dragonfly adult males and their conspecific larvae at various spatial scales in CFR rivers (Chapter 3). Both life stages are renowned bioindicators, but little is known about how well adults represent the larvae, and whether freshwater assessments should be based on one or both life stages. I expect conspecific adults and larvae to occur close to each other, and to be influenced by the same environmental variables. This would be owing to the territorial nature of adult male dragonflies, combined with the responsibility to select the best conditions to ensure the survival of their larvae. However, I expect this response to be dependent on scale of sampling sites, with larger scales showing the highest levels of overlap.

Thereafter, I utilize male adult dragonflies as bioindicators and assess the success of two conservation initiatives applied to transformed landscapes in the KwaZulu-Natal Province. It is necessary to determine the value of approaches aimed at reducing the negative impacts associated with transformation in order to provide recommendations for current management and future implementation. I used dragonflies to determine whether EN corridors in a forestry

setting (Chapter 4) and a conservation pond within an urban environment (Chapter 5) were useful biodiversity conservation methods for these transformed landscapes. I expect both of these approaches to be successful for conserving dragonflies in transformed landscapes, and dragonflies to be good indicators of their successes. Furthermore, I think the importance of correctly managing and creating heterogeneity within these settings will be evident, as seen for other taxa. Each of these chapters have been prepared as separate entities intended for journal publicaition, thus some repetition may be unavoidable. These chapters are set out below (title and major objectives provided):

Chapter 2: **Aquatic insects act as umbrella species for biodiversity in rivers of the biodiversity rich Cape Floristic Region**

1. Evaluate the performance of the umbrella species approach for dragonflies and EPT taxa, using the umbrella index, as a tool for biodiversity conservation.
2. Determine whether dragonflies and EPT species respond similarly to environmental variables as both groups are sensitive to changes in their habitats.

Chapter 3: **Does father know best? Exploring the links between adult male dragonflies and conspecific larvae**

1. Determine whether conspecific larvae and male adult dragonflies occur within close proximity to each other - ‘father knows best’ hypothesis.
2. Test the associations between larvae and adults at three spatial scales.
3. Determine whether larvae and adults are driven by the same environmental conditions - ‘shared environmental preferences’ hypothesis.

Chapter 4: **Well-designed large-scale conservation ecological networks maintain river ecosystem integrity and dragonfly diversity in forestry plantations**

1. Compare dragonfly diversity between rivers within timber ENs to nearby untransformed rivers in PAs surrounded by natural grassland.
2. Test the influence of corridor width on dragonfly diversity.
3. Define the environmental drivers of dragonfly diversity within ENs and PAs.

4. Determine which dragonfly functional and life-history traits are being selected for by the environments of the ENs and PAs.

Chapter 5: **Lessons learnt from thirty years of monitoring a pond designed for dragonfly conservation**

1. Assess the responses of dragonflies to changes in habitat heterogeneity due to pond biotope creation, biological succession due to lack of maintenance, and recovery after pond restoration

2. Utilize dragonflies to monitor pond restoration.

3. Highlight the important variables to consider in order to maintain high dragonfly diversity at urban conservation ponds

Chapter 6: **General conclusions**

1.7 References

Alberti, M., Booth, D., Hill, K., Coburn, B., Avolio, C., Coe, S., & Spirandelli, D. (2007). The impact of urban patterns on aquatic ecosystems: An empirical analysis in Puget lowland sub–basins. Landscape and Urban Planning, 80(4), 345–361.

Angelibert, S., & Giani, N. (2003). Dispersal characteristics of three odonate species in a patchy habitat. Ecography, 26(1), 13–20.

Baker, S. C., & Sharp Jr, H. F. (1998). Evaluation of the recovery of a polluted urban stream using the Ephemeroptera-Plecoptera-Trichoptera index. Journal of Freshwater Ecology, 13(2), 229 – 234.

Bennett, E. M., Carpenter, S. R., & Caraco, N. F. (2001). Human impact on erodable phosphorus and eutrophication: A global perspective increasing accumulation of phosphorus in soil threatens rivers, lakes, and coastal oceans with eutrophication. BioScience, 51(3), 227–234.

Betrus, C. J., Fleishman, E., & Blair, R. B. (2005). Cross–taxonomic potential and spatial transferability of an umbrella species index. Journal of Environmental Management, 74(1), 79–87.

Bird, M. S., Mlambo, M. C., Wasserman, R. J., Dalu, T., Holland, A. J., Day, J. A., Villet, M. H., Bilton, D. T., Barber–James, H. M., & Brendonck, L. (2019). Deeper knowledge of shallow waters: reviewing the invertebrate fauna of southern African temporary wetlands. Hydrobiologia, 827(1), 89–121.

Bockstaller, C., & Girardin, P. (2003). How to validate environmental indicators. Agricultural Systems, 76(2), 639–653.

Bonada, N., Prat, N., Resh, V. H., & Statzner, B. (2006). Developments in aquatic insect biomonitoring: A comparative analysis of recent approaches. Annual Review of Entomology, 51(1), 495–523.

Boon, P., & Raven, P. (2012). River conservation and management. John Wiley and Sons, Chichester, UK.

Boulton, A. J. (1999). An overview of river health assessment: philosophies, practice, problems and prognosis. Freshwater Biology, 41(2), 469–479.

Bried, J. T., D'amico, F., & Samways, M. J. (2012). A critique of the dragonfly delusion hypothesis: why sampling exuviae does not avoid bias. Insect Conservation and Diversity, 5(5), 398–402.

Bried, J. T., Dillon, A. M., Hager, B. J., Patten, M. A., & Luttbeg, B. (2015a). Criteria to infer local species residency in standardized adult dragonfly surveys. Freshwater Science, 34(3), 1105 – 1113.

Bried, J. T., Hassall, C., Simaika, J. P., Corser, J. D., & Ware, J. (2015). Directions in dragonfly applied ecology and conservation science. Freshwater science, 34(3), 1020 – 1022.

Bried, J. T., Herman, B. D., & Ervin, G. N. (2007). Umbrella potential of plants and dragonflies for wetland conservation: A quantitative case study using the umbrella index. Journal of Applied Ecology, 44(4), 833–842.

Briers, R. A., & Biggs, J. (2003). Indicator taxa for the conservation of pond invertebrate diversity. Aquatic Conservation: Marine and Freshwater Ecosystems, 13(4), 323–330.

Brodin, T. (2008). Behavioral syndrome over the boundaries of life—carryovers from larvae to adult damselfly. Behavioral Ecology, 20(1), 30 – 37.

Bulánková, E. (1997). Dragonflies (Odonata) as bioindicators. Biologia, Bratislava, 52(2), 177 – 180.

Buskirk, R. E., & Sherman, K. J. (1985). The influence of larval ecology on oviposition and mating strategies in dragonflies. Florida Entomologist, 39 – 51.

Cairns, J., McCormick, P. V., & Niederlehner, B. R. (1993). A proposed framework for developing indicators of ecosystem health. Hydrobiologia, 263(1), 1 – 44.

Ceballos, G., Ehrlich, P. R., & Dirzo, R. (2017). Biological annihilation via the ongoing sixth mass extinction signalled by vertebrate population losses and declines. Proceedings of the National Academy of Sciences, 114(30), E6089–E6096.

Ceballos G., García A., & Ehrlich P. R. (2010). The sixth extinction crisis: Loss of animal populations and species. Journal of Cosmology, 8, 1821–1831.

Chovanec, A. (2000). Dragonflies (Insecta: Odonata) as indicators of the ecological integrity of aquatic systems–a new assessment approach. Internationale Vereinigung für theoretische und angewandte Limnologie: Verhandlungen, 27(2), 887 – 890.

Chutter, F. M. (1995). The role of aquatic organisms in for sustainable utilisation. Water Science and Technology, 32(5–6), 283–291.

Conrad, K. F., Willson, K. H., Harvey, I. F., Thomas, C. J., & Sherratt, T. N. (1999). Dispersal characteristics of seven odonate species in an agricultural landscape. Ecography, 22(5), 524–531.

Corbet, P. S. (1980). Biology of Odonata. Annual review of entomology, 25(1), 189 – 217.

Corbet, P. S. (1999). Dragonflies: behaviour and ecology of Odonata. Harley books.

Dabrowski, J. M., Peall, S. K. C., Reinecke, A. J., Liess, M., & Schulz, R. (2002). Runoff–related pesticide input into the Lourens River, South Africa: basic data for exposure assessment and risk mitigation at the catchment scale. Water, Air, and Soil Pollution, 135(1–4), 265–283.

Dale, V. H., & Beyeler, S. C. (2001). Challenges in the development and use of ecological indicators. Ecological Indicators, 1(1), 3–10.

Dallas, H. F. (2004). Seasonal variability of macroinvertebrate assemblages in two regions of South Africa: implications for aquatic bioassessment. African Journal of Aquatic Science, 29(2), 173–184.

Dallas, H. F. (2013). Ecological status assessment in Mediterranean rivers: complexities and challenges in developing tools for assessing ecological status and defining reference conditions. Hydrobiologia, 719(1), 483–507.

Dallas, H. F., & Day, J. A. (2007). Natural variation in macroinvertebrate assemblages and the development of a biological banding system for interpreting bioassessment data – A preliminary evaluation using data from upland sites in the south–western Cape, South Africa. Hydrobiologia, 575(1), 231–244.

Darwall, W. R. T., Smith, K. G., Allen, D., Seddon, M. B., Reid, G. M., Clausnitzer, V., & Kalkman, V. J. (2009). Freshwater biodiversity – a hidden resource under threat. In wildlife in a changing world: an analysis of the 2008 IUCN Red List of Threatened Species. IUCN (pp. 43 – 54), Gland, Switzerland.

Day, J., & Day, B. (2009). The endemic aquatic invertebrate fauna of the Cape Floristic Realm: what does the future hold? In IOP Conference Series: Earth and Environmental Science. IOP Publishing (Vol. 6, No. 31, p. 312015).

De Wit, M., Behrendt, H., Bendoricchio, G., & Bleuten, W. (2002). The contribution of agriculture to nutrient pollution in three European rivers, with reference to the European Nitrates Directive. European Water Management Online, 1–19.

Dickens, C. W. S., & Graham, P. M. (2002). The South African scoring system (SASS) version 5 rapid bioassessment method for rivers. African Journal of Aquatic Science, 27(1), 1–10.

Dijkstra, K. D. B., Monaghan, M. T., & Pauls, S. U. (2014). Freshwater biodiversity and aquatic insect diversification. Annual Review of Entomology, 59, 143–163.

D'Amico, F., Darblade, S., Avignon, S., Blanc-Manel, S., & Ormerod, S. J. (2004). Odonates as indicators of shallow lake restoration by liming: comparing adult and larval responses. Restoration Ecology, 12(3), 439 – 446.

Dudgeon, D., Arthington, A. H., Gessner, M. O., Kawabata, Z. I., Knowler, D. J., Lévêque, C., Naiman, R. J., Prieur–Richard, A. H., Soto, D., Stiassny, M. L., & Sullivan, C. A. (2006).

Freshwater biodiversity: importance, threats, status and conservation challenges. Biological Reviews, 81(2), 163–182.

Duelli, P., & Obrist, M. K. (2003). Biodiversity indicators: the choice of values and measures. Agriculture, Ecosystems and Environment, 98(1–3), 87–98.

Dye, P., & Versfeld, D. (2007). Managing the hydrological impacts of South African plantation forests: An overview. Forest Ecology and Management, 251(1–2), 121–128.

Edwards, M. B. P., & Roberts, P. J. T. (2006). Managing forests for water: the South African experience. International Forestry Review, 8(1), 65–71.

Fairbanks, D. H., Hughes, C. J., & Turpie, J. K. (2004). Potential impact of viticulture expansion on habitat types in the Cape Floristic Region, South Africa. Biodiversity and Conservation, 13(6), 1075–1100.

Fleishman, E., Blair, R. B., & Murphy, D. D. (2010). Empirical validation of a method for umbrella species selection. Ecological Applications, 11(5), 1489–1501.

Fleishman, E., Murphy, D., & Brussard, P. F. (2000). A new method for selection of umbrella species for conservation planning. Ecological Applications, 10(2), 569–579.

Frankel, O. H., & Soulé, M. E. (1981). Conservation and evolution. Cambridge University Press, Cambridge, United Kingdom.

Frazee, S. R., Cowling, R. M., Pressey, R. L., Turpie, J. K., & Lindenberg, N. (2003). Estimating the costs of conserving a biodiversity hotspot: a case–study of the Cape Floristic Region, South Africa. Biological Conservation, 112(1–2), 275–290.

Futahashi, R., Kawahara–Miki, R., Kinoshita, M., Yoshitake, K., Yajima, S., Arikawa, K., & Fukatsu, T. (2015). Extraordinary diversity of visual opsin genes in dragonflies. Proceedings of the National Academy of Sciences, 112(11), E1247–E1256.

Gaylard, A., Owen–Smith, N., & Redfern, J. (2003). Surface water availability: implications for heterogeneity and ecosystem processes. In the Kruger experience: ecology and management of savanna heterogeneity. Island Press (pp. 171 – 188), Washington, USA.

Gerhardt, A. (2002). Bioindicator species and their use in biomonitoring. Environmental Monitoring, 1, 77–123.

Gerten, D., Rost, S., von Bloh, W., & Lucht, W. (2008). Causes of change in 20th century global river discharge. Geophysical Research Letters, 35(20).

Gledhill, D. G., James, P., & Davies, D. H. (2005). Urban pond: A landscape of multiple meanings. Paper presented at the 5th International Postgraduate Research Conference in The Built and Human Environment. University of Salford, Manchester, UK.

Goldblatt, P., & Manning, J. C. (1999). Cape flora – a conspectus of the Cape Flora of South Africa. National Botanical Institute, Pretoria, South Africa.

Graham, N. (1999). Urban water. In guidelines for drinking–water quality, 2nd edition. World Health Organisation, Geneva, Switzerland.

Green, P. A., Vörösmarty, C. J., Harrison, I., Farrell, T., Sáenz, L., & Fekete, B. M. (2015). Freshwater ecosystem services supporting humans: Pivoting from water crisis to water solutions. Global Environmental Change, 34, 108–118.

Giugliano, L., Hardersen, S., & Santini, G. (2012). Odonata communities in retrodunal ponds: a comparison of sampling methods. International Journal of Odonatology, 15(1), 13 – 23.

Habel, J. C., Samways, M. J., & Schmitt, T. (2019). Mitigating the precipitous decline of terrestrial European insects: Requirements for a new strategy. Biodiversity and Conservation, 28(6), 1343–1360.

Haddad, N. M. (1999). Corridor and distance effects on interpatch movements: a landscape experiment with butterflies. Ecological Applications, 9(2), 612–622.

Haddad, N. M., Bowne, D. R., Cunningham, A., Danielson, B. J., Levey, D. J., Sargent, S., & Spira, T. (2003). Corridor use by diverse taxa. Ecology, 84(3), 609–615.

Harabiš, F., & Dolný, A. (2012). Human altered ecosystems: Suitable habitats as well as ecological traps for dragonflies (Odonata): The matter of scale. Journal of Insect Conservation, 16(1), 121–130.

Hardersen, S. (2008). Dragonfly (Odonata) communities at three lotic sites with different hydrological characteristics. Italian Journal of Zoology, 75, 271–283.

Harrison, I. J., Abell, R., Darwall, W., Thieme, M. L., Tickner, D., & Timboe, I. (2018). The freshwater biodiversity crisis. Science, 362(6421), 1369–1369.

Harrison, I. J., Green, P. A., Farrell, T. A., Juffe–Bignoli, D., Sáenz, L., & Vörösmarty, C. J. (2016). Protected areas and freshwater provisioning: a global assessment of freshwater provision, threats and management strategies to support human water security. Aquatic Conservation: Marine and Freshwater Ecosystems, 26, 103–120.

Hassall, C. (2014). The ecology and biodiversity of urban ponds. Wiley Interdisciplinary Reviews: Water, 1(2), 187–206.

Hassall, C., & Anderson, S. (2015). Stormwater ponds can contain comparable biodiversity to unmanaged wetlands in urban areas. Hydrobiologia, 745(1), 137–149.

Hawking, J. H., & New, T. R. (1999). The distribution patterns of dragonflies (Insecta: Odonata) along the Kiewa River, Australia, and their relevance in conservation assessment. Hydrobiologia, 392(2), 249–260.

Hawking, J. H., & New, T. R. (2002). Interpreting dragonfly diversity to aid in conservation assessment: lessons from the Odonata assemblage at Middle Creek, north-eastern Victoria, Australia. Journal of Insect Conservation, 6(3), 171 – 178.

Hornung, J. P., & Rice, C. L. (2003). Odonata and wetland quality in southern Alberta, Canada: a preliminary study. Odonatologica, 32(2), 119–129.

Hurley, B. P., Croft, P., Verleur, M., Wingfield, M. J., & Slippers, B. (2012). The control of the *Sirex* woodwasp in diverse environments: the South African experience. In The *Sirex* Woodwasp and its Fungal Symbiont. Springer (pp. 247–264), Dordrecht, the Netherlands.

Jackson, L. E., Kurtz, J. C., & Fisher, W. S. (2000). Evaluation Guidelines for Ecological Indicators. US Environmental Protection Agency, Washington, USA.

Joubert, L., & Samways, M. J. (2014). Equivalence of grasslands in an ecological network and a World Heritage Site. Biodiversity and Conservation, 23(10), 2415–2426.

Kemp, D. J. (2018). Habitat selection and territoriality. In insect behavior: from mechanisms to ecological and evolutionary consequences. Oxford University Press (pp. 80 – 93), Oxford, UK.

Kietzka, G. J., Pryke, J. S., & Samways, M. J. (2015). Landscape ecological networks are successful in supporting a diverse dragonfly assemblage. Insect Conservation and Diversity, 8(3), 229–237.

Lee Foote, A., & Rice Hornung, C. L. (2005). Odonates as biological indicators of grazing effects on Canadian prairie wetlands. Ecological Entomology, 30(3), 273–283.

Le Maitre, D. C., Van Wilgen, B. W., Chapman, R. A., & McKelly, D. H. (1996). Invasive plants and water resources in the Western Cape Province, South Africa: modelling the consequences of a lack of management. Journal of Applied Ecology, 161 – 172.

Lenat, D. R., & Barbour, M. T. (1994). Using benthic macroinvertebrate community structure for rapid, cost-effective, water quality monitoring: rapid bioassessment. Biological monitoring of aquatic systems. Lewis Publishers, Boca Raton, Florida, 187 – 215.

Lipsey, M. K., & Hockey, P. A. R. (2010). Do ecological networks in South African commercial forests benefit grassland birds? A case study of a pine plantation in KwaZulu–Natal. Agriculture, Ecosystems and Environment, 137(1–2), 133–142.

Lubertazzi, M. A. A., & Ginsberg, H. S. (2009). Persistence of dragonfly exuviae on vegetation and rock substrates. Northeastern Naturalist, 16(1), 141 – 148.

Macadam, C. R., & Stockan, J. A. (2015). More than just fish food: ecosystem services provided by freshwater insects. Ecological Entomology, 40, 113–123.

Magoba, R. N., & Samways, M. J. (2010). Recovery of benthic macroinvertebrate and adult dragonfly assemblages in response to large scale removal of riparian invasive alien trees. Journal of Insect Conservation, 14(6), 627–636.

Mallin, M. A., Ensign, S. H., Wheeler, T. L., & Mayes, D. B. (2002). Pollutant removal efficacy of three wet detention ponds. Journal of Environmental Quality, 31(2), 654–660.

Mancini, L., Formichetti, P., Anselmo, A., Tancioni, L., Marchini, S., & Sorace, A. (2005). Biological quality of running waters in protected areas: the influence of size and land use. Biodiversity and Conservation, 14, 351–364.

Masese, F. O., & Raburu, P. O. (2017). Improving the performance of the EPT Index to accommodate multiple stressors in Afrotropical streams. African Journal of Aquatic Science, 42(3), 219–233.

May, C. W., Horner, R. R., Karr, J. R., Mar, B. W., & Welch, E. B. (1997). Effects of urbanization on small streams in the Puget Sound lowland ecoregion. Watershed Protection Techniques, 2(4), 483–494.

Maynou, X., Martin, R., & Aranda, D. (2017). The role of small secondary biotopes in a highly fragmented landscape as habitat and connectivity providers for dragonflies (Insecta: Odonata). Journal of Insect Conservation, 21(3), 517–530.

McCauley, S. J. (2007). The role of local and regional processes in structuring larval dragonfly distributions across habitat gradients. Oikos, 116(1), 121–133.

McGeogh, M. A. (1998). The selection, testing and application of terrestrial insects as bioindicators. Biological Reviews, 73(2), 181–201.

McGeoch, M. A., & Chown, S. L. (1998). Scaling up the value of bioindicators. Trends in Ecology & Evolution, 13(2), 46 – 47.

Moore, N. W. (1957). Territory in dragonflies and birds. Bird study, 4(3), 125 – 130.

Murphy, H. T., & Lovett-Doust, J. (2004). Context and connectivity in plant metapopulations and landscape mosaics: does the matrix matter? Oikos, 105(1), 3–14.

Neke, K. S., & Du Plessis, M. A. (2004). The threat of transformation: quantifying the vulnerability of grasslands in South Africa. Conservation Biology, 18(2), 466–477.

Nel, J. L., Reyers, B., Roux, D. J., & Cowling, R. M. (2009). Expanding protected areas beyond their terrestrial comfort zone: identifying spatial options for river conservation. Biological Conservation, 142(8), 1605–1616.

Nõges, P., Argillier, C., Borja, Á., Garmendia, J. M., Hanganu, J., Kodeš, V., Pletterbauer, F., Sagouis, A., & Birk, S. (2016). Quantified biotic and abiotic responses to multiple stress in freshwater, marine and ground waters. Science of the Total Environment, 540, 43–52.

Noss, R. F. (1990). Indicators for monitoring biodiversity: a hierarchical approach. Conservation Biology, 4(4), 355–364.

Patten, M. A., Bried, J. T., & Smith-Patten, B. D. (2015). Survey data matter: predicted niche of adult vs breeding Odonata. Freshwater Science, 34(3), 1114 – 1122.

Patten, M. A., Hjalmarson, E. A., Smith-Patten, B. D., & Bried, J. T. (2019). Breeding thresholds in opportunistic Odonata records. Ecological Indicators, 106, 105460.

Prendergast, J. R., & Eversham, B. C. (1997). Species richness covariance in higher taxa: empirical tests of the biodiversity indicator concept. Ecography, 20(2), 210–216.

Primack, R., Kobori, H., & Mori, S. (2000). Dragonfly pond restoration promotes conservation awareness in Japan. Conservation Biology, 14(5), 1553–1554.

Pryke, S. R., & Samways, M. J. (2001). Width of grassland linkages for the conservation of butterflies in South African afforested areas. Biological Conservation, 101(1), 85–96.

Pryke, J. S., & Samways, M. J. (2012a). Ecological networks act as extensions of protected areas for arthropod biodiversity conservation. Journal of Applied Ecology, 49(3), 591–600.

Pryke J. S. & Samways, M .J. (2012b). Conservation management of complex natural forest and plantation edge effects. Landscape Ecology, 27, 73–85.

Pryke, J. S., & Samways, M. J. (2015). Conserving natural heterogeneity is crucial for designing effective ecological networks. Landscape Ecology, 30(4), 595–607.

Pryke, J. S., Samways, M. J., & De Saedeleer, K. (2015). An ecological network is as good as a major protected area for conserving dragonflies. Biological Conservation, 191, 537–545.

Raebel, E. M., Merckx, T., Riordan, P., Macdonald, D. W., & Thompson, D. J. (2010). The dragonfly delusion: why it is essential to sample exuviae to avoid biased surveys. Journal of Insect Conservation, 14(5), 523–533.

Remsburg, A. (2011). Relative influence of prior life stages and habitat variables on dragonfly (Odonata: Gomphidae) densities among lake sites. Diversity, 3(2), 200 – 216.

Revenga, C., Campbell, I., Abell, R., De Villiers, P., & Bryer, M. (2005). Prospects for monitoring freshwater ecosystems towards the 2010 targets. Philosophical Transactions of the Royal Society B: Biological Sciences, 360(1454), 397–413.

Richardson, D. M. (1998). Forestry trees as invasive aliens. Conservation biology, 12(1), 18-26.

Richardson, D. M., & Van Wilgen, B. W. (2004). Invasive alien plants in South Africa: how well do we understand the ecological impacts?: working for water. South African Journal of Science, 100(1–2), 45–52.

Roberge, J. M., & Angelstam, P. E. R. (2004). Usefulness of the umbrella species concept as a conservation tool. Conservation Biology, 18(1), 76–85.

Rouget, M., Richardson, D. M., & Cowling, R. M. (2003). The current configuration of protected areas in the Cape Floristic Region, South Africa—reservation bias and representation of biodiversity patterns and processes. Biological Conservation, 112(1–2), 129–145.

Russi, D., ten Brink, P., Farmer, A., Badura, T., Coates, D., Förster, J., Kumar, R., & Davidson, N. (2013). The economics of ecosystems and biodiversity for water and wetlands. IEEP, London and Brussels, UK and Belgium.

Samways, M. J. (1992). Dragonfly conservation in South Africa: a biogeographical perspective. Odonatologica, 21(2), 165–180.

Samways, M. J. (2008). Dragonflies and damselflies of South Africa (No. 70). Pensoft Publishers, Sophia, Bulgaria.

Samways, M. J., Bazelet, C. S. & Pryke, J. S. (2010). Provision of ecosystem services by large scale corridors and ecological networks. Biodiversity and Conservation, 19, 2949–2962.

Samways, M. J., & Pryke, J. S. (2016). Large–scale ecological networks do work in an ecologically complex biodiversity hotspot. Ambio, 45(2), 161–172.

Samways, M. J., & Sharratt, N. J. (2010). Recovery of endemic dragonflies after removal of invasive alien trees. Conservation Biology, 24(1), 267–277.

Samways, M. J., & Simaika, J. P. (2016). Manual of freshwater assessment: Dragonfly biotic index. South African Biodiversity Institute, Pretoria, South Africa.

Samways, M. J., & Taylor, S. (2004). Impacts of invasive alien plants on Red–Listed South African dragonflies (Odonata): working for water. South African Journal of Science, 100(1–2), 78–80.

Samways, M. J., Taylor, S., & Tarboton, W. (2005). Extinction reprieve following alien removal. Conservation Biology, 19(4), 1329–1330.

Sánchez–Bayo, F., & Wyckhuys, K. A. (2019). Worldwide decline of the entomofauna: A review of its drivers. Biological Conservation, 232, 8–27.

Saunders, D. J., Meeuwig, J. J., & Vincent, C. J. (2002). Freshwater protected areas: strategies for conservation. Conservation Biology, 16, 30–41.

Schindler, M., Fesl, C., & Chovanec, A. (2003). Dragonfly associations (Insecta: Odonata) in relation to habitat variables: a multivariate approach. Hydrobiologia, 497(1–3), 169–180.

Sih, A., Bell, A. M., Johnson, J. C., & Ziemba, R. E. (2004). Behavioral syndromes: an integrative overview. The quarterly review of biology, 79(3), 241-277.

Simaika, J. P., & Samways, M. J. (2009). An easy–to–use index of ecological integrity for prioritizing freshwater sites and for assessing habitat quality. Biodiversity and Conservation, 18(5), 1171–1185.

Simaika, J. P., & Samways, M. J. (2011). Comparative assessment of indices of freshwater habitat conditions using different invertebrate taxon sets. Ecological Indicators, 11, 370–378.

Simaika, J. P., & Samways, M. J. (2012). Using dragonflies to monitor and prioritize lotic systems: a South African perspective. Organisms Diversity and Evolution, 12(3), 251–259.

Simaika, J. P., Samways, M. J., & Frenzel, P. P. (2016). Artificial ponds increase local dragonfly diversity in a global biodiversity hotspot. Biodiversity and Conservation, 25(10), 1921–1935.

Smith, J., Samways, M. J., & Taylor, S. (2007). Assessing riparian quality using two complementary sets of bioindicators. Biodiversity and Conservation, 16(9), 2695–2713.

Spellerberg, I. F. (1991). Monitoring Ecological Change. Cambridge University Press, Cambridge, UK.

Steytler, N. S., & Samways, M. J. (1995). Biotope selection by adult male dragonflies (Odonata) at an artificial lake created for insect conservation in South Africa. Biological Conservation, 72(3), 381–386.

Stoks, R., & Cordoba–Aguilar, A. (2012). Evolutionary ecology of Odonata: a complex life cycle perspective. Annual Review of Entomology, 57, 249–265.

Stoks, R., Debecker, S., Van, K. D., & Janssens, L. (2015). Integrating ecology and evolution in aquatic toxicology: insights from damselflies. Freshwater Science, 34(3), 1032 – 1039.

Strayer, D. (2006). Challenges for freshwater invertebrate conservation. Journal of the North American Benthological Society, 25(2), 271–287.

Suh, A. N., & Samways, M. J. (2001). Development of a dragonfly awareness trail in an African botanical garden. Biological Conservation, 100(3), 345–353.

Suh, A. N., & Samways, M. J. (2005). Significance of temporal changes when designing a reservoir for conservation of dragonfly diversity. Biodiversity and Conservation, 14(1), 165–178.

Taylor, L., & Hochuli, D. F. (2017). Defining greenspace: Multiple uses across multiple disciplines. Landscape and Urban Planning, 158, 25–38.

Trombulak, S. C., & Frissell, C. A. (2000). Review of ecological effects of roads on terrestrial and aquatic communities. Conservation Biology, 14(1), 18–30.

UNEP. (2016). A snapshot of the world's water quality: towards a global assessment. United Nations Environment Programme (UNEP), Nairobi, Kenya.

Valente-Neto, F., de Oliveira Roque, F., Rodrigues, M. E., Juen, L., & Swan, C. M. (2016). Toward a practical use of Neotropical odonates as bioindicators: Testing congruence across taxonomic resolution and life stages. Ecological Indicators, 61, 952 – 959.

Vörösmarty, C. J., McIntyre, P. B., Gessner, M. O., Dudgeon, D., Prusevich, A., Green, P., Glidden, S., Bunn, S.E., Sullivan, C.A., Liermann, C.R., & Davies, P. M. (2010). Global threats to human water security and river biodiversity. Nature, 467(7315), 555.

Wasson, R. J. (1996). Land use and climate impacts on fluvial systems during the period of agriculture. In PAGES workshop report, series (pp. 96-2).

Wendel, H. E. W., Downs, J. A., & Mihelcic, J. R. (2011). Assessing equitable access to urban green space: the role of engineered water infrastructure. Environmental Science and Technology, 45(16), 6728–6734.

Wishart, M. J., & Day, J. A. (2002). Endemism in the freshwater fauna of the south–western Cape, South Africa. Limnologie, 28(4), 1–5.

Woodward, G., Perkins, D. M., & Brown, L. E. (2010). Climate change and freshwater ecosystems: impacts across multiple levels of organization. Philosophical Transactions of the Royal Society B: Biological Sciences, 365(1549), 2093–2106.

Chapter 2: Does father know best? Exploring the links between adult male dragonflies and conspecific larvae

Abstract

Dragonflies are globally renowned bioindicators, with both larval and adult life stages used in freshwater quality assessments. Despite this, little is known about how well adults represent the larvae, and what this means for freshwater assessments based on one life stage. Here, I investigate whether dragonfly larvae and adults have similar responses to biotic and abiotic factors. Firstly, I test the hypothesis that adult male dragonflies are likely to occur in close proximity to their conspecific aquatic larvae ('father knows best' hypothesis). I test this possibility at three different spatial scales. Secondly, I test the hypothesis that larvae and adults share similar responses to environmental variables ('shared environmental preferences' hypothesis). At medium and large spatial scales, larvae and adults strongly correlated for abundance, species richness, Shannon diversity, and Dragonfly Biotic Index (DBI) scores. However, at the small spatial scale, only 15% of observations matched (contained adults and conspecific larvae). This increased to 46% at the medium scale, and 60% at the large scale, neither of which were significantly different from the number of mismatches, thus refuting the 'father knows best' hypothesis. Water temperature correlated with agricultural land use, and were the only variables dictating similar responses by larvae and adults. Aside from these, larvae and adult assemblages were driven by different variables, which is not as expected under the 'shared environmental preferences' hypothesis. Despite the rejection of both hypotheses, it is evident that the type of question investigated determines the appropriate scale for surveys, and whether or not the life stages can be used interchangeably. Where large stretches of river are surveyed, for coarse ecological questions, either larvae or adults can be sampled. However, for biodiversity surveys using fine ecological scales such as pools, riffles or glides, both larvae and adults should be sampled to give comprehensive results, as recommended in the DBI manual.

2.1 Introduction

Freshwater insects occupy many types of habitats, and are highly sensitive to environmental change, making them useful for making conservation management decisions (Brito et al., 2018). They can be used in part for determining site prioritization for conservation action, and as sensitive indicators, can rapidly detect potential environmental problems, as well as being useful for monitoring change over time (Shafie et al., 2017; Morrison et al., 2018). They are relatively low-cost and easy to sample (Brito et al., 2018), and their small body sizes, global abundance and short life cycles add to their value as indicators (Agouridis et al., 2015). Despite these advantages, the taxonomic challenge inhibits use of some species in these assessments (Allan & Flecker, 1993). Others, such as the Odonata (encompassing dragonflies and damselflies but hereafter referred to collectively as dragonflies), are well-studied taxonomically, and are globally recognised for their indicator potential (Renner et al., 2016; Samways & Simaika, 2016; de Moor, 2017; Lesch & Bouwman, 2018).

The hemimetabolous life cycle of dragonflies, as with many aquatic insects, is characterised by an extreme realm shift, from an aquatic insect to a terrestrial one (Stoks & Cordoba-Aguilar, 2012). The aquatic larval stage is the longest in duration, often lasting months, and in some species up to several years. Larvae are predatory, grow rapidly, and moult many times. For the final moult, larvae leave their aquatic habitats and metamorphose into the aerial, adult phase. Young adults disperse away from water bodies, and once sexually mature, return to suitable aquatic habitats in their reproductive phase (Corbet, 1999; Stoks & Cordoba-Aguilar, 2012).

Many adult male dragonflies are territorial, and defend their carefully selected habitats from other conspecifics to select a mate with which to reproduce within this territory (Kemp, 2018). In principle then, both the adult males and their conspecific larvae should therefore occur at the same general sites. Although both the larval and adult stages are used in freshwater assessments, the most suitable stage is dependent on various factors: the specific research or management questions being asked, geographical area being surveyed, time of year, and degree of knowledge of the dragonflies in the area.

Globally, records of dragonflies typically favour the adult over the larval stage (Kalkman et al., 2018). For example, out of 1 410 055 records from five European countries (Belgium, France,

Netherlands, Slovenia and UK), 85% to 98% are adults, and 0.06 % to 15 % comprise larvae (Raebel et al., 2010). Compared to the larvae, adults are conspicuous and more appealing to people (particularly the colourful males) and are easier to sample and identify to species level (Lemelin, 2007; Clausnitzer et al., 2017). However, in cooler climates, they can only be surveyed comprehensively during the warmer months (when the adult phase is present), and when conditions are sunny and windless (Kietzka et al., 2018; McCauley, Hammond & Mabry, 2018). Owing to their high mobility, adults easily disperse to areas where they do not necessarily reproduce. These areas may act as stepping-stone, temporary habitats, favoured for food resources rather than as breeding habitats (Simaika et al., 2016; Maynou et al., 2017). Therefore, some authors have criticized use of adults in assessments, as they may lead to unreliable overestimates of reproductive populations (Hardersen, 2008; Raebel et al., 2010).

Residency can be assumed for adult Odonata species observed in mating habitat. This is because they generally copulate and oviposit only in viable freshwater habitats but only recording these individuals would be a severe underrepresentation. Exuviae and tenerals are sure indications of reproductive success and life cycle completion in an area (Raebel et al., 2010). However, tenerals are difficult to identify and to handle without damaging them and will disperse within a few hourse once their bodies have hardened (Patten et al., 2015). Exuviae, don't persist for a long period of time after emergence, especially in areas faced with adverse weather conditions (Lubertazzi et al., 2009). In contrast to adults, larval surveys can take place under most weather conditions. However, in-water sampling techniques require greater effort compared to visual adult sampling, which can also accurately cover a larger area. Conditions such as fast-flowing water, dangerous animals like crocodiles, and harmful diseases such as bilhazia can inhibit aquatic sampling (Samways, 2008; Bird et al., 2019). The probability of detecting larvae is usually also lower than that of adults, due to their cryptic nature, and species-specific micro-habitat requirements (Niba & Samways 2006; Bried et al., 2012). Additionally, in many countries around the world, the larvae of many species have not yet been not described, and for many species, knowledge of habitat preferences is crucial in order to find them (Samways, 1992). Even for described species, larval identification may be difficult, time-consuming, and requires a high level of expertise. The season of sampling is also important, because for accurate identification, individuals should be in their final-instar. These challenges may result in under-representation of the actual assemblage when only using larvae (Simaika & Samways, 2009a; 2011; Briggs et al., 2019).

Despite these various challenges, both life stages are used as indicators of water quality, and in the development of indices. In South Africa, the Dragonfly Biotic Index (DBI) uses the presence of dragonfly adults, supplemented by larvae and exuviae when available, as a measure of ecological integrity based on the total complement of species present in a defined area (Simaika & Samways, 2009a; 2012; Samways & Simaika, 2016). As it operates at the species level, it is a highly sensitive index (Simaika & Samways, 2009a), and functions very favourably in comparison with many other indices (Rosset et al., 2013). It has been used successfully to measure habitat recovery, selection of sites for conservation action, and in assessments of stream restoration after the removal of alien trees species (Samways & Taylor, 2004; Simaika & Samways, 2011; 2012; Harabiš & Dolný, 2012).

Other indices use higher-level taxa of benthic macroinvertebrates to assess ecological integrity, with the aim of providing an indication of water quality (Dickens & Graham, 1998; 2002). Dragonfly larvae have been incorporated into some of these assessments, such as the South African Scoring System (SASS), which is a major part of the National River Health Programme (Uys et. al.,1996; Dickens & Graham, 2002). There is a strong correlation between adult dragonfly scores (DBI) and macroinvertebrate scores (SASS) (Dickens & Graham, 2002; Smith et. al., 2007; Simaika & Samways, 2009a), but whether this correlation extends to the composition of dragonfly larvae at the species level is unknown.

Biotope preferences are important to consider for conservation of dragonflies, and for their use in assessments as indicators (Clark & Samways, 1996; Suh & Samways, 2001). Vegetation-related characteristics are central drivers of adult dragonfly assemblages (Samways & Taylor, 2004; Samways & Sharratt, 2010). In rivers of the Cape Florisitc Region (CFR) South Africa, alien tree canopies shade dragonfly habitats, and have a major effect on adults (Samways & Sharratt, 2010). Other environmental conditions that influence their assemblages include in-water parameters (Kietzka, Pryke & Samways, 2017), flow rate, shade, water permanency (Clark & Samways, 1996), aquatic macrophytes, and vegetation height (Dunkle, 1976; Corbet, 1999; Samways & Taylor, 2004; Samways & Sharratt, 2010), as well as elevation (Samways, 1989; Samways & Steytler, 1996; Hawking & New, 1999; Clausnitzer, 2003).

In reservoirs in South Africa, larvae are driven by aquatic and marginal vegetation, elevation, water body depth, pH, turbidity and water temperature (Niba & Samways, 2006). Often, species have their own specific biotopes or features that they favour or require for survival (Steytler & Samways, 1995; Stewart & Samways, 1998). Some adult dragonflies, such as, *Nesciothemis farinosa* and *Crocothemis erythraea*, favour sunny conditions for thermoregulation, whereas *Notiothemis jonesi* and *Chlorolestes tessellatus* prefer shaded habitats with sunflecks (Osborn & Samways, 1996). Some larvae, especially burrowing species, are influenced by substrate characteristics (Huggins & DuBois, 1982). The South African gomphid, *Paragomphus cognatus*, prefers coarse sand for burrowing, and avoids fine sand habitats (Keetsch & Moran, 1966).

Although adult dragonflies are not directly dependent on the aquatic environment, their larvae are. Osborn and Samways (1996) and Kietzkaet al. (2017) suggest that perhaps adult dragonflies select habitats based on certain features required by their larvae. The larva is the longest developmental stage, and selecting sites with suitable conditions for complete larval development would seem highly adaptive. Therefore, adult dragonflies and conspecific larvae may occur at the same sites, and respond similarly to environmental variables. In support of this, Kietzka et al. (2017) found that for natural sections of rivers in the CFR, in-water parameters were the most important drivers of adult dragonfly assemblages, even though they likely do not directly depend on these. Also, some species such as *Orthetrum caffrum* and *O. chrysostigma* characteristically perch on sticks or vegetation overhanging mud, which may be linked to the mud-dwelling habits of their larvae (Osborn & Samways, 1996). Alternatively, species that breed in isolated habitats, like certain CFR rivers, may remain close to the sites from where they emerged (Corbet, 1999; Samways & Simaika, 2016), which would strengthen the link between adult and larval habitat co-occurrence, and responses to the same environmental variables. This was the case for ponds in the UK, where the majority of individuals remained at their natal ponds (Conrad et al., 1999). Despite these suggestions, the spatial and environmental connections between dragonfly adults and conspecific larva have not yet been investigated in detail at various spatial scales.

The CFR is a renowned biodiversity hotspot, not only for being rich in endemic plant species but also for its exceptionally high levels of endemic, aquatic invertebrates (Wishart & Day, 2002). The mosaic structure of CFR rivers creates a range of diverse habitats to colonize (Meek et al., 2010). Around two thirds of the aquatic invertebrates are endemic to the CFR region, and represent more

than a third of South Africa's aquatic invertebrate species (Wishart & Day, 2002). The mountainous rivers are also a major centre of endemism for dragonfly species (Simaika & Samways, 2009b). The heterogeneous nature of these rivers, coupled with the high habitat specificities of the many unique, endemic dragonfly species, make it a suitable area to determine whether there are links between habitats (and environmental factors) of conspecific dragonfly larvae and adults. Here, I aim to determine whether conspecific larvae and adults occur close to each other ('father knows best' hypothesis). In doing so, it is important to consider the influence of spatial scale, especially for dragonflies, owing to their variously high mobility. Therefore, I test the associations between the larvae and adults at three spatial scales, to help select the appropriate scale for future assessments. Then I determine whether larvae and adults are influenced by the same environmental conditions ('shared environmental preferences' hypothesis). Due to the territorial nature of many adult male dragonflies, coupled with selection pressure to choose the best-suited conditions to ensure the survival of their progeny, I expect to find larvae and adults of the same species close to each other, and to be driven by the same environmental variables.

2.2 Materials and methods

2.2.1 Study area and sites

Study sites were along four rivers in the Western Cape Province of South Africa (the Eerste (34°00'28.19'S; 19°00'6.46'E), Lourens (34°00'56.90'S; 18°58'42.54'E), Molenaars (33°42'00'S; 19°13'60'E) and Palmiet (34°03'24.95'S; 19°00'48.52'E) Rivers). All rivers had untransformed sections of natural fynbos vegetation, as well as sections of cultivated land, planted with vineyards and orchards. Untransformed sites of the Eerste, Lourens and Molenaars Rivers were in the upper river reaches, while agricultural sites were downstream (middle reaches). However, in the case of the Palmiet River, the untransformed sites were in the lower reaches, and agricultural sites in the upper reaches. The choice of a range of variously disturbed habitats was to 'stretch' the responses of the two life stages under conditions from transformed to fully historic.

Ninety sites were randomly selected per river using QGIS 2.14.3 (QGIS Development Team, 2014). These were divided between the natural and agricultural land uses. Each site comprised a

10 x 3 m area of water, starting from the river's edge. Sampling took place twice per site, once during spring/early summer and again in late summer/autumn, to account for both larvae and adult species seasonal differences. Sites were evenly distributed between three flow regimes: still deposition pools (< 0.01 m/sec), gently flowing glides (surface flow 0.01 - 0.4 m/sec), and fast flowing riffles (surface flow > 0.4 m/sec).

2.2.2 Sampling of adult, exuviae and larval dragonflies

On warm, cloudless days, two observers recorded the species and the abundance of mature male dragonflies present at each site. Observations were conducted for 30 min/site, using close-focus binoculars, between 10:00 and 15:00. Observers used a butterfly net to collect voucher specimens or individuals that were difficult to identify in the field. These were later identified according to Samways and Simaika (2016). The species *Trithemis dorsalis* and *T. furva* are indistinguishable in the field, and were recorded as one taxon ('TD'). Exuviae were searched for along the same transects used for adults for 30min/site.

Aquatic larvae were sampled along the same transects used for adult observations. River substrate was disturbed by foot, and with a 30 x 30 cm, 1 000 μm mesh net, which simultaneously collected the suspended samples. To account for habitat differences between species, all microhabitats present at each site were sampled for flow rates, vegetation, and substrate. Each site was sampled for 3 minutes, after which, larvae were separated from debris on a tray, and preserved in 90% ethanol. Individuals close to or in their final instar were identified to species according to Suhling et al. (2014). Specimens that were too small to accurately identify were excluded from the dataset.

2.2.3 Environmental variables

At each site, 24 environmental variables (EVs) were recorded. These included the categorical variables of river catchment (Eerste, Lourens, Palmiet, or Molenaars Rivers), land use (0 = natural, 1 = agricultural), flow type (pool, glide, or riffle). Percentage shade was visually estimated, and river width and depth at each site was determined with a tape measure. A handheld GPS recorded

elevation and site positions. A multi-parameter water quality meter (Model: YSI 556 Multi Probe System; Make: YSI Fondriest Environmental) was used to measure water parameters (temperature, dissolved oxygen, conductivity, and pH) at each site. For both river edges and beds, two observers visually estimated percentages of rocks, sand, and detritus. For vegetation data, average vegetation height, as well as the percentage cover, of both alien and indigenous plant structures were estimated per site. Percentage cover of *Prionium serratum* (Palmiet Reed) and aquatic macrophytes were recorded as separate variables, as they have a strong influence on dragonflies (Samways & Sharratt, 2010). Water temperature and agricultural land use were strongly positively correlated, and so land use was excluded from analyses.

2.2.4 Data analyses

2.2.4.1 Analyses for the 'father knows best' hypothesis

To determine whether sampling effort was sufficient, the non-parametric species estimators ICE, Chao2, and second-order Jackknife were calculated in R 3.5.2 (R Core Team, 2018) using the fossil package (Vavrek, 2011). For insect assemblages where a large number of rare species occur, as in the CFR, non-parametric species estimators are recommended (Hortal et al., 2006). Rarefied species accumulation curves were constructed for the larvae, excuviae and adult dragonflies in R (R Core Team, 2018) using the vegan package (Oksanen et al., 2015). There were such poor returns from surveys of exuviae, that they were not included in any further analyses.

Three datasets were used for all analyses, and included larvae, adults, and physically adjusted adults. The adjusted adult dataset excluded six species (*Zosteraeschna minuscula*, *Chlorolestes umbratus*, *Ceriagrion glabrum*, *Tramea limbata*, *Pantala flavescens*, and *Elatoneura glauca*) that were only observed as adults, and not as larvae.

Whether the relationships between dragonfly larvae and adults are scale dependent is unknown, and therefore three different scales were used in analyses. The small scale comprised 360 sites of 10 x 3 m (90 per river), with each site at least 50 m from the nearest site. At the medium scale, there were 40 sites and each covered an area of 90 x 3 m. Here, for each site, data were pooled from nine adjacent small scale sites (10 sites per river). The large scale had eight sites and each

covered an area of 450 x 3 m (2 per river). Here, for each site, data were pooled from five adjacent medium scale sites (2 sites per river).

Analyses were run using four assemblage measures: abundance, species richness, Shannon diversity index scores, and Dragonfly Biotic Index scores (DBI). The *vegan* package (Oksanen et al., 2015) in R (R Core team, 2018) was used to calculate Shannon diversity index scores per site. For each observed species, DBI scores were obtained from Samways and Simaika (2016). Thereafter, the average score per site was calculated by dividing the sum of all the species' DBI scores at a site by the number of species observed at that site.

Spearman rank correlations in R (R Core Team, 2018) tested the degree of association between larvae vs. adults, and larvae vs. adjusted adults for abundance, species richness, Shannon diversity index scores, and DBI scores at the three spatial scales.

To determine whether larvae and adults of the same species occurred at the same sites, exact tests of goodness-of-fit were to be performed with the *binom.test* function in the native *stats* package in R (R Core Team, 2018). The hypothesis tested was that there would be significantly different proportions of matches and mismatches between the larvae and adults of a species, against the null hypothesis that there would be no differences, assuming an equal likelihood of both. The constituents of the function are the number of successes/matches (the number of matches between the same species of larvae and adult), the number of trials: number of matches + number of mismatches (sites where only the larvae or the adult of a species was found), and the hypothesized probability of success (here 0.5). Here, the exact test was selected over a chi-squared or G-test, as it is applicable to sample sizes of < 1 000 or when at least one of the expected values is small (five or less). Analyses were run at the three spatial scales, for all the species, and sites combined, and then for each species individually. Bar graphs of species proportions were plotted with *ggplot2* (Wickham & Chang, 2008). Similarly, a bar graph of larvae and adult abundances per species was also plotted.

2.2.4.2 Analyses for the 'shared environmental preferences' hypothesis

Further analyses aimed to determine whether larvae and adults were driven by the same environmental variables. For dragonfly abundance, species richness, Shannon index, and DBI scores, best fit models were determined for larvae, adults, and adjusted adults, with a stepwise forward selection method based on AIC values using the *AICcmodavg* package (Mazerolle & Mazerolle, 2017) in R (R Core Team, 2018). Because all data were found to be non-normal in the *lme4* package (Bates et al., 2014), generalized linear mixed-effect models (GLMMs) were run. As data were not over-dispersed, the GLMMs could accurately identify which environmental factors significantly influenced the four response variables of the three groups (larvae, adults, and adjusted adults). River identity, as well as elevation, were included as the random effects. Abundance and species richness data best fitted a Poisson distribution (Bolker et al., 2009), and Shannon and DBI score data were transformed using the formula: x = (x - minimum value)/(maximum value - minimum value), thereafter they best fitted a binomial distribution.

To select the groups of the most important environmental variables that influenced the larvae, adults, and adjusted adults assemblage structure, Canonical Correspondence Analyses (CCAs) were run in CANOCO 5 (ter Braak & Šmilauer, 2012). This was done separately for each assemblage using interactive forward selection analyses and 9999 permutations (ter Braak, 1990). In the *mvabund* package (Wang et al., 2012) in R (R Core Team, 2018), the function *manyglm* determined the best fit distribution for the data. Thereafter, GLMs with multivariate extensions were run with abundance data for larvae, adults and adjusted adults using the variables selected by the CCAs. These were best-fitted with a negative binomial distribution, which assumes a quadratic mean-variance. This model-based approach, tests multiple effects at the assemblage level, by fitting a separate GLM to each species, using the given explanatory variables. This method accounts for between-species correlations, and uses resampling-based hypothesis testing to determine which environmental variables are associated with the abundances (Wang et al., 2012).

2.3 Results

2.3.1 Results for the 'father knows best' hypothesis

A total of 1 795 larvae (19 species) and 1 268 adult (25 species) dragonfly individuals were sampled. Both rarefaction curves neared asymptote, which indicates that sampling effort was

sufficient (Fig. 2.1). This was further supported by species estimates for larvae (ICE = 19.37, Chao2 = 19.17 and second-order Jackknife = 18) and adults (ICE = 27.42, Chao2 = 30 and second-order Jackknife = 27) nearing observed species richness.

Only 118 exuviae of only five species were discovered, which is too low a number and species richness to be included for statistical analyses, and far from the recorded number of larvae or adult species. The rarefaction curve quickly reached asymptote, which means for this method increased sampling effort would not have increased the number of species observed (Fig. 2.1). No exuviae at all were recorded along the agricultural section of the Palmiet River, the natural section of the Eerste River, or the entire Molenaars River.

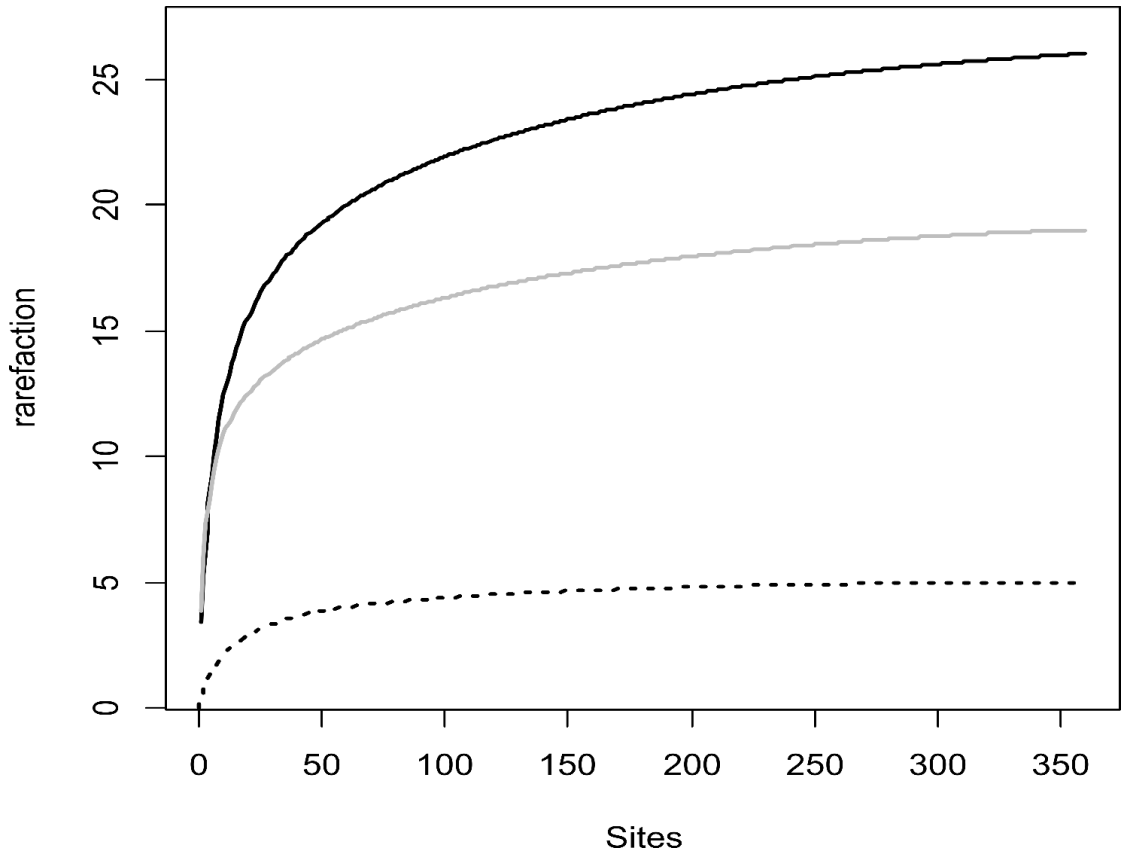

Fig. 2.1 Rarefied species accumulation curves for dragonflies. Solid black line = adults, solid grey line = larvae, and dashed black line = exuviae.

2.3.1.1 Larval and adult associations

At the small scale, there were positive but weak correlations between larvae and adults, as well as for larvae and adjusted adults for abundance, species richness, Shannon index, and DBI scores (Table 2.1; Appendix 2.A; 2.B). Correlations for all four assemblage measures strengthened as spatial scale increased for larvae vs. adults, and larvae vs. adjusted adults, with the strongest correlations at the largest scale (Table 2.1). Despite the high R_s values at the large spatial scale, the P-values for Shannon diversity and DBI scores were not significant at the 5% level large scale, where sample sizes were small (Table 2.1).

Exact tests analysed the proportions of matches vs. mismatches, and at the small scale, there were 179 matches (proportion: 0.15) vs. 1 024 mismatches (proportion: 0.85), with a significant difference in the distribution skewed towards the higher proportion of mismatches (Table 2.2; Fig. 2.2), meaning that the observed proportions differed from the expected proportions (0.5: 0.5). Seventeen of the species had significantly different proportions of matches vs. mismatches, skewed towards the mismatches (Table 2.2; Fig. 2.2). Eight species (*Chlorolestes conspicuus* (CC), *Ceratogomphus pictus* (CP), *Elattoneura frenulata* (EF), *Syncordulia gracilis* (SG), *Elattoneura glauca* (EG), *Pantala flavescens* (PFL), *Tramea limbata* (TL) and *Zosteraeschna minuscula* (ZM)) did not have significantly different proportions of matches vs. mismatches, although the proportions of mismatches were always higher (Table 2.2; Fig. 2.2). Of these eight species, half (EG, PFL, TL and ZM) were only observed as adults, and never as larvae. At the medium scale, 157 matches (proportion: 0.46) vs. 186 mismatches (proportion: 0.54) were observed, and did not differ significantly (Table 2.2). For the species individually, *Allocnemis leucosticta* (AL), *Anax speratus* (AS), *Crocothemis erythraea* (CE), *C. sanguinolenta* (CS), *Pinheyschna subpupillata* (PS), and *Trithemis dorsalis/furva* (TD) had significantly different proportions of matches vs. mismatches (Table 2.2; Fig. 2.3). All of the abovementioned species, except for *P. subpupillata* (PS), had significantly greater proportions of mismatches. At the large scale, 55 matches (proportion: 0.60) vs. 36 mismatches (proportion: 0.40) were observed (Fig. 2.4). Although skewed towards the proportion of matches, this was not significant (Table 2.2). Again, *P. subpupillata* (PS) was the only species with a significant difference between proportions of matches vs. mismatches, skewed towards matches (Fig. 2.4). Although not always significant, as scale increased, a greater proportion of matches occurred. At the small scale, there were no species with higher proportions of matches than mismatches, and this increased to five species at

the medium scale, while reaching 12 species at the large scale (Fig. 2.2; 2.3; 2.4). As scale increased, so did the probability of success for all the species (Table 2.2). In terms of abundance per species, eight species had a greater number of larvae than adults, and the remaining 17 species had higher adult abundance than larvae (Fig. 2.5).

Table 2.1 Spearman rank correlations for larvae vs. adults and larvae vs. statistically-adjusted adults for dragonfly abundance, species richness, Shannon diversity index scores, and Dragonfly Biotic Index scores. Bolded numbers signify significance. R_s = Spearman's Rank Correlation Coefficient, DBI = Dragonfly Biotic Index.

		Larvae			
Small scale		**Abundance**	**Richness**	**Shannon**	**DBI**
Adults	R_s	0.32	0.34	0.31	0.13
	p-value	**< 0.001**	**< 0.001**	**< 0.001**	**0.017**
Adjusted adults	R_s	0.33	0.34	0.30	0.11
	p-value	**< 0.001**	**< 0.001**	**< 0.001**	**0.046**
Medium scale					
Adults	R_s	0.61	0.65	0.48	0.65
	p-value	**< 0.001**	**< 0.001**	**0.002**	**< 0.001**
Adjusted adults	R_s	0.62	0.70	0.55	0.62
	p-value	**< 0.001**	**< 0.001**	**< 0.001**	**< 0.001**
Large scale					
Adults	R_s	0.79	0.89	0.60	0.67
	p-value	**0.028**	**0.003**	0.132	0.082
Adjusted adults	R_s	0.78	0.90	0.55	0.67
	p-value	**0.023**	**0.003**	0.171	0.069

Table 2.2 Exact tests results of proportions between matches vs mismatches. POS = probability of success. Bolded numbers signify significance. See Appendix 2.C for species names.

Species	Small scale		Medium scale		Large scale	
	p-value	POS	p-value	POS	p-value	POS
Overall	< **0.001**	0.13	0.130	0.46	0.059	0.60
AI	< **0.001**	0.09	0.424	0.40	1.000	0.50
AL	< **0.001**	0.00	**0.003**	0.08	0.625	0.25
AS	< **0.001**	0.00	**0.001**	0.11	0.688	0.33
CC	0.250	0.00	0.250	0.00	1.000	1.00
CE	< **0.001**	0.00	**0.008**	0.00	0.125	0.00
CG	**0.029**	0.27	0.625	0.75	1.000	1.00
CP	0.500	0.00	0.500	0.00	1.000	0.00
CS	< **0.001**	0.00	**0.002**	0.00	0.375	0.20
CU	**0.021**	0.10	0.375	0.20	1.000	1.00
EF	0.063	0.00	0.125	0.00	0.250	0.00
EG	0.125	0.00	0.250	0.00	0.250	0.00
OJ	< **0.001**	0.11	0.875	0.48	0.727	0.63
PFL	< **0.001**	0.09	0.375	0.80	1.000	1.00
PC	< **0.001**	0.10	0.108	0.68	0.219	0.83
PD	< **0.001**	0.13	1.000	0.48	0.289	0.75
PF	< **0.001**	0.18	0.524	0.41	1.000	0.50
PFI	1.000	0.00	1.000	0.00	1.000	0.00
PK	< **0.001**	0.13	0.405	0.39	1.000	0.57
PS	< **0.001**	0.27	< **0.001**	0.95	**0.008**	1.00
SG	1.000	0.00	1.000	0.00	1.000	0.00
TA	< **0.001**	0.19	0.834	0.46	0.688	0.67
TD	< **0.001**	0.04	**0.004**	0.25	0.727	0.38
TL	0.250	0.00	0.250	0.00	1.000	1.00
ZM	0.125	0.14	0.375	0.20	1.000	1.00
ZN	< **0.001**	0.14	0.791	0.57	0.625	0.75

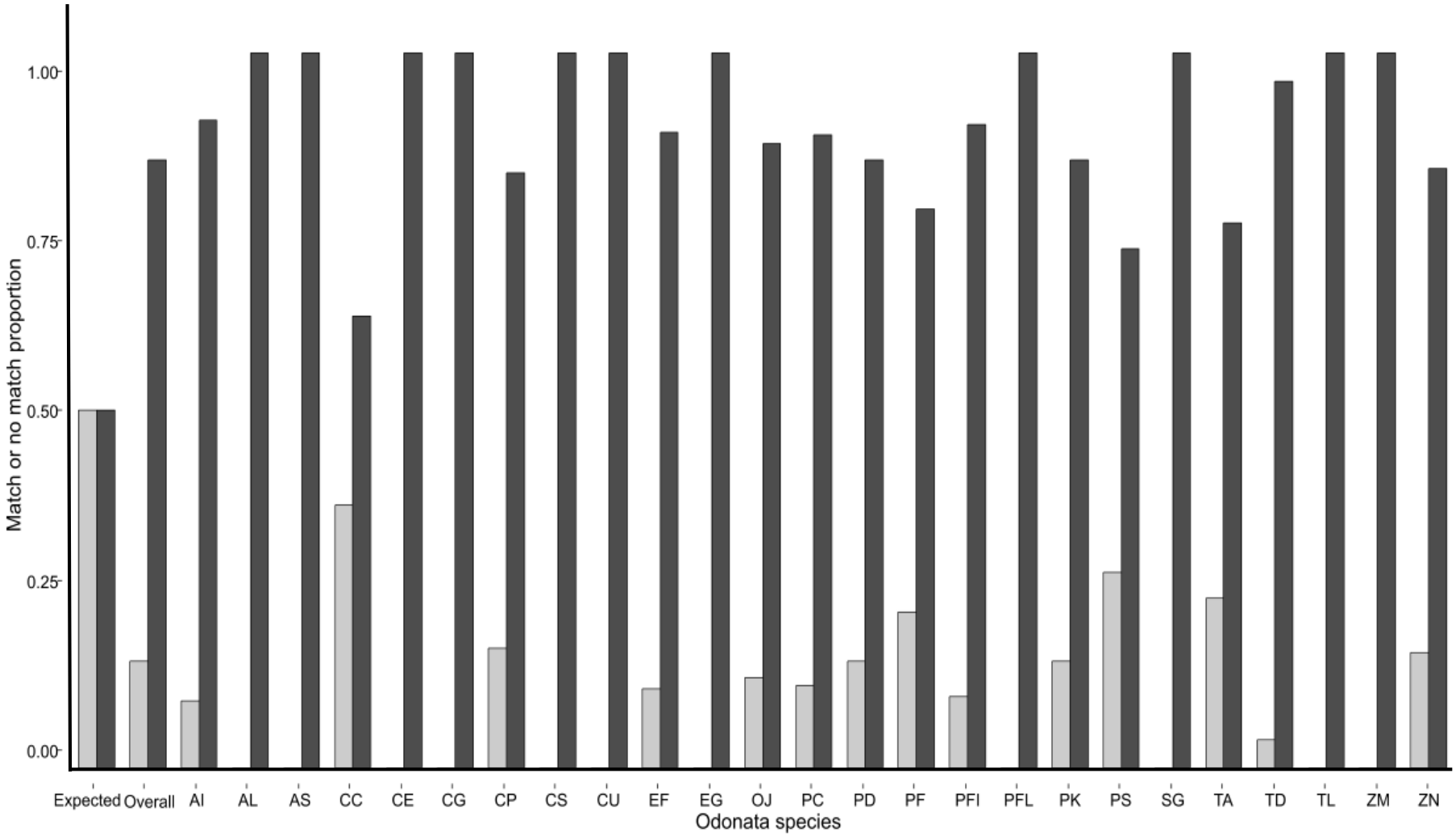

Fig. 2.2 Small scale proportions of matches (grey) and mismatches (black) for overall dragonflies, and each species separately. See Appendix 2.C for species names.

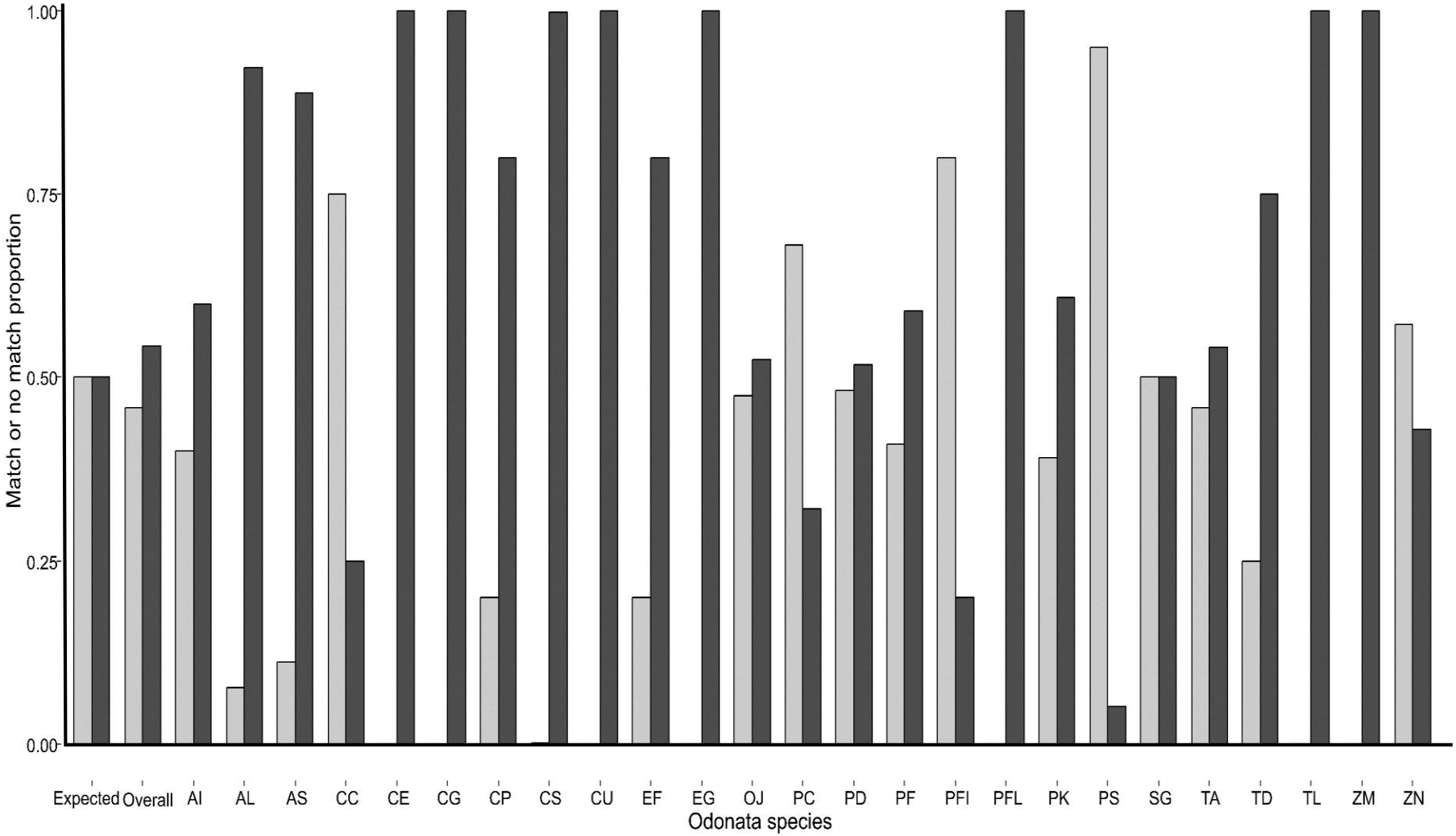

Fig. 2.3 Medium scale proportions of matches (grey) and mismatches (black) for overall dragonflies and each species separately. See Appendix 2.C for species names.

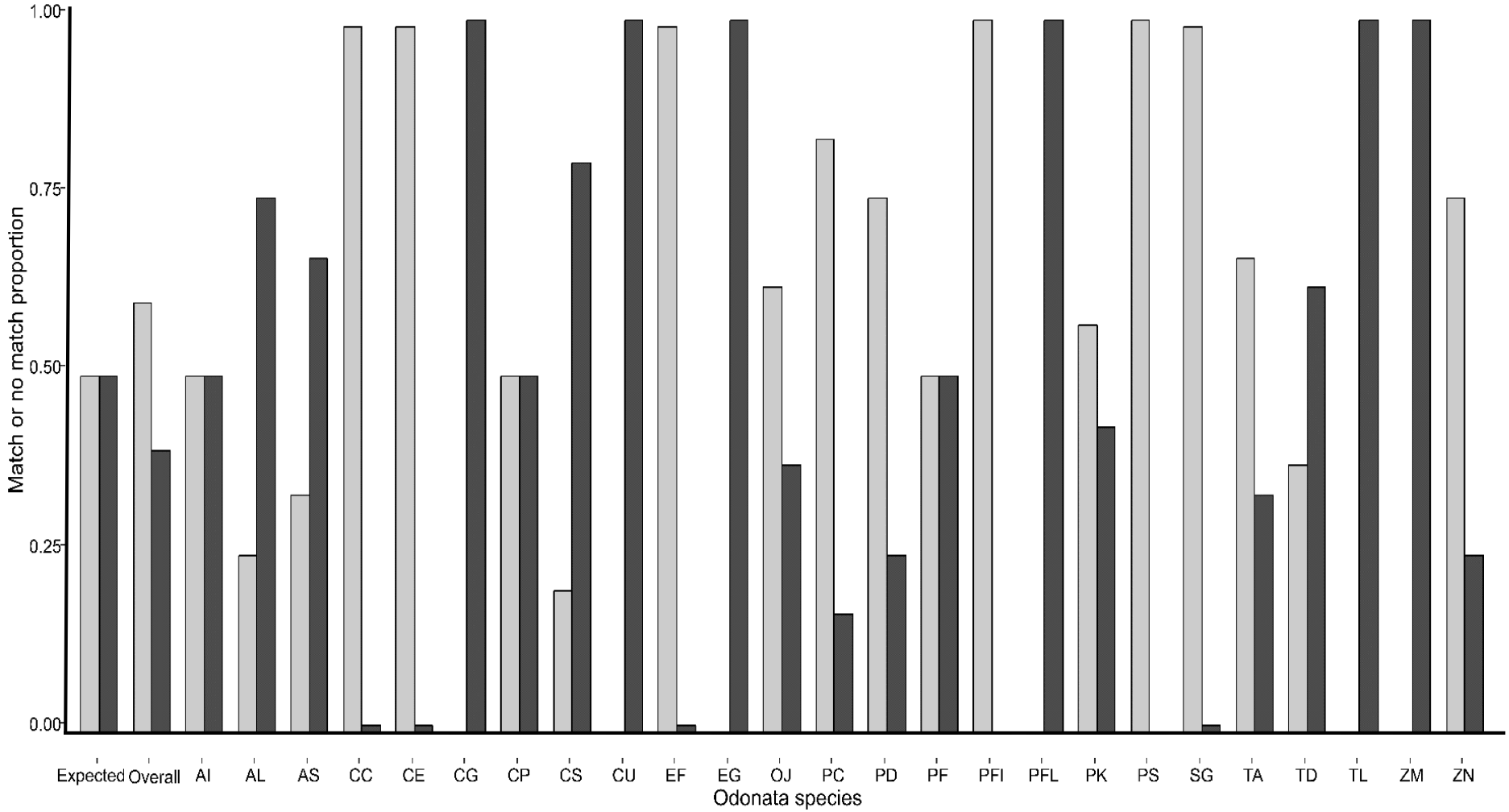

Fig. 2.4 Large scale proportions of matches (grey) and mismatches (black) for overall dragonflies and each species separately. See Appendix 2.C for species names.

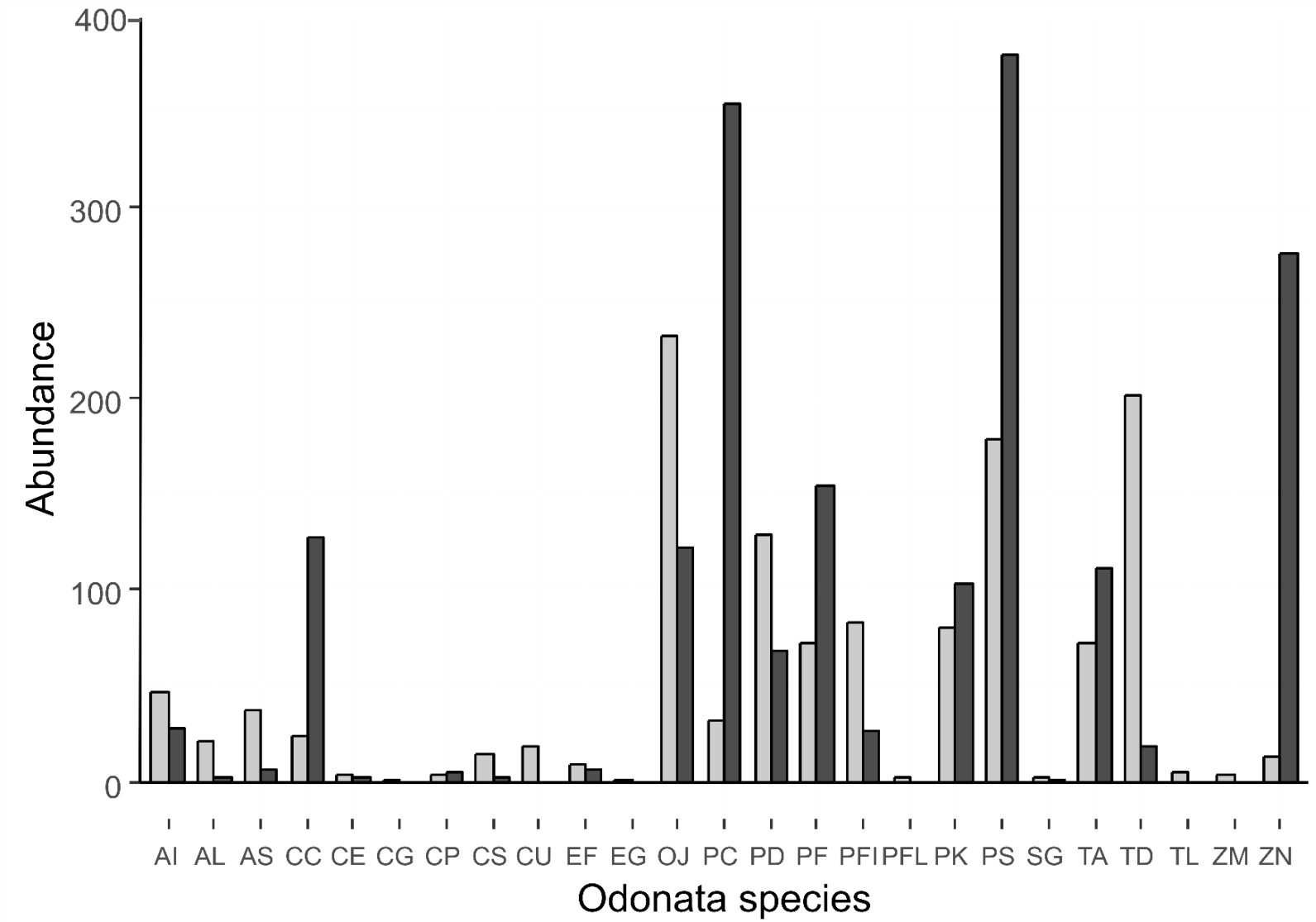

Fig. 2.5 Overall larvae and adult abundances, grey = adult, black = larvae. See Appendix 2.C for species names.

2.3.2 Results for 'shared environmental preferences' hypothesis

Water temperature was the only environmental variable that significantly influenced larvae, adults, and adjusted adults for all four of the assemblage parameters (Fig. 2.6; 2.7). Abundance, species richness, and Shannon diversity scores positively correlated with water temperature, whereas DBI scores negatively correlated (Fig. 2.6; 2.7). Larval abundance and species richness were significantly lower in fast-flowing water (riffles). River width positively correlated with larval abundance, but negatively correlated with adult DBI scores. Adult abundance, species richness, and adjusted adult abundance all positively correlated with river depth (Fig. 2.6). In terms of water parameters, there were positive correlations between larval abundance and conductivity, and for larval Shannon diversity scores and dissolved oxygen (Fig. 2.6; 2.7). Both larval Shannon diversity and DBI scores negatively correlated with pH (Fig. 2.7). Species richness and Shannon diversity scores for the larvae negatively correlated with the percentage of sand covering the riverbed, whereas adult abundance, adjusted adult abundance, and species richness positively correlated with sand cover (Fig. 2.6; 2.7). Larval DBI scores positively correlated with percentage shade. Abundance, species richness, and Shannon scores for the adults and adjusted adults positively correlated with the percentage of *P. serratum* reed and aquatic macrophytes. Whereasadult DBI scores negatively correlated with aquatic macrophyte cover, average vegetation height negatively correlated with adult and adjusted adult abundance, while positively correlating with adult DBI scores (Fig. 2.6; 2.7).

Water temperature had the greatest influence on species composition of the larvae, adults, and adjusted adults. An additional six environmental variables were significant for all three of the groups (Table 2.3). These included flow type, shade cover, river width, pH, and percentage rocks on the side of the river and on the riverbed. Only for the larvae was assemblage composition significantly affected by percentage cover of debris and sand on the riverbed. Composition of the adults and adjusted adults were significantly influenced by *P. serratum* cover and aquatic macrophytes.

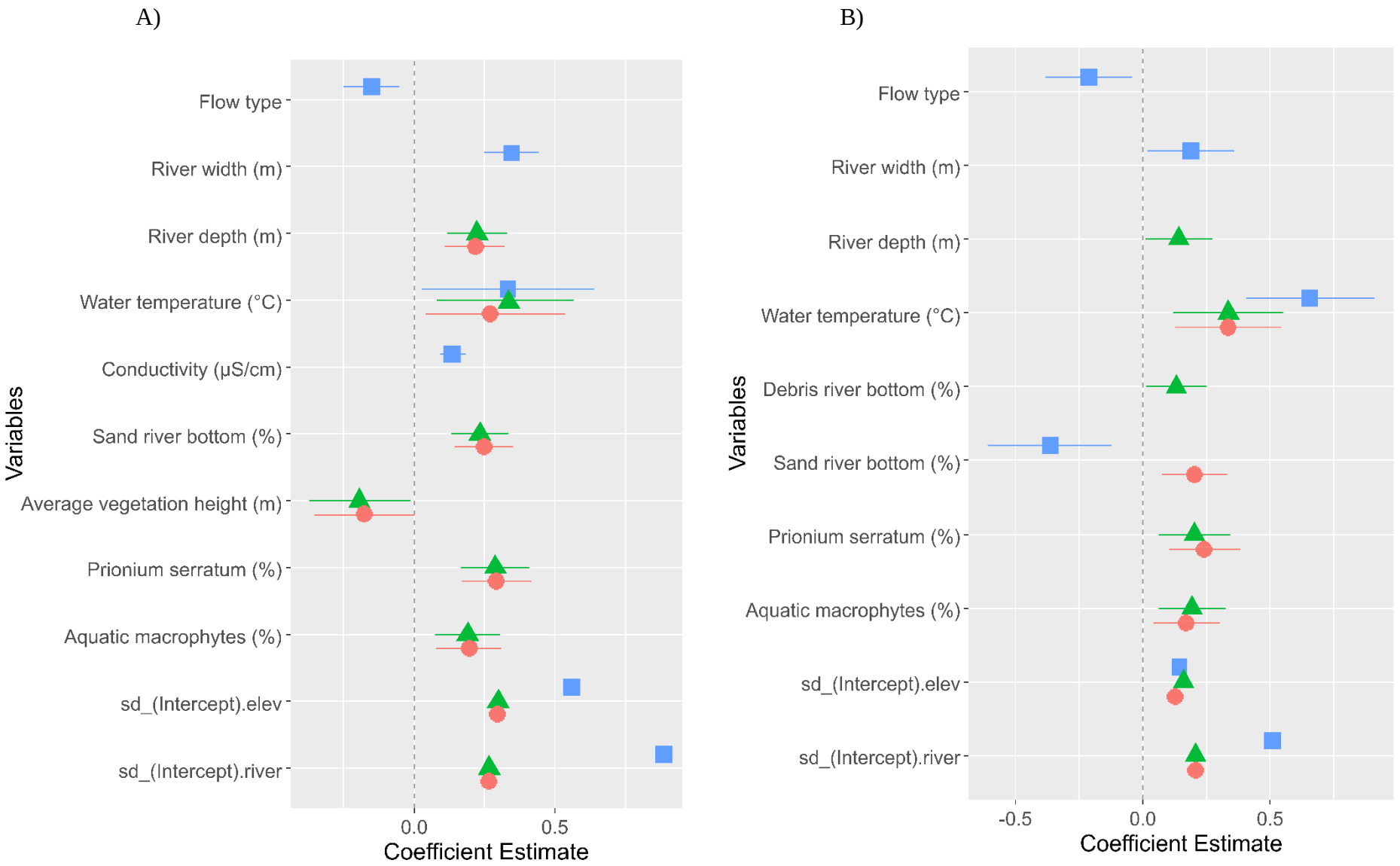

Fig. 2.6 Significant environmental variables influencing A) abundance, and B) species richness.

Squares = larvae; triangles = adults; circle = adjusted adults.

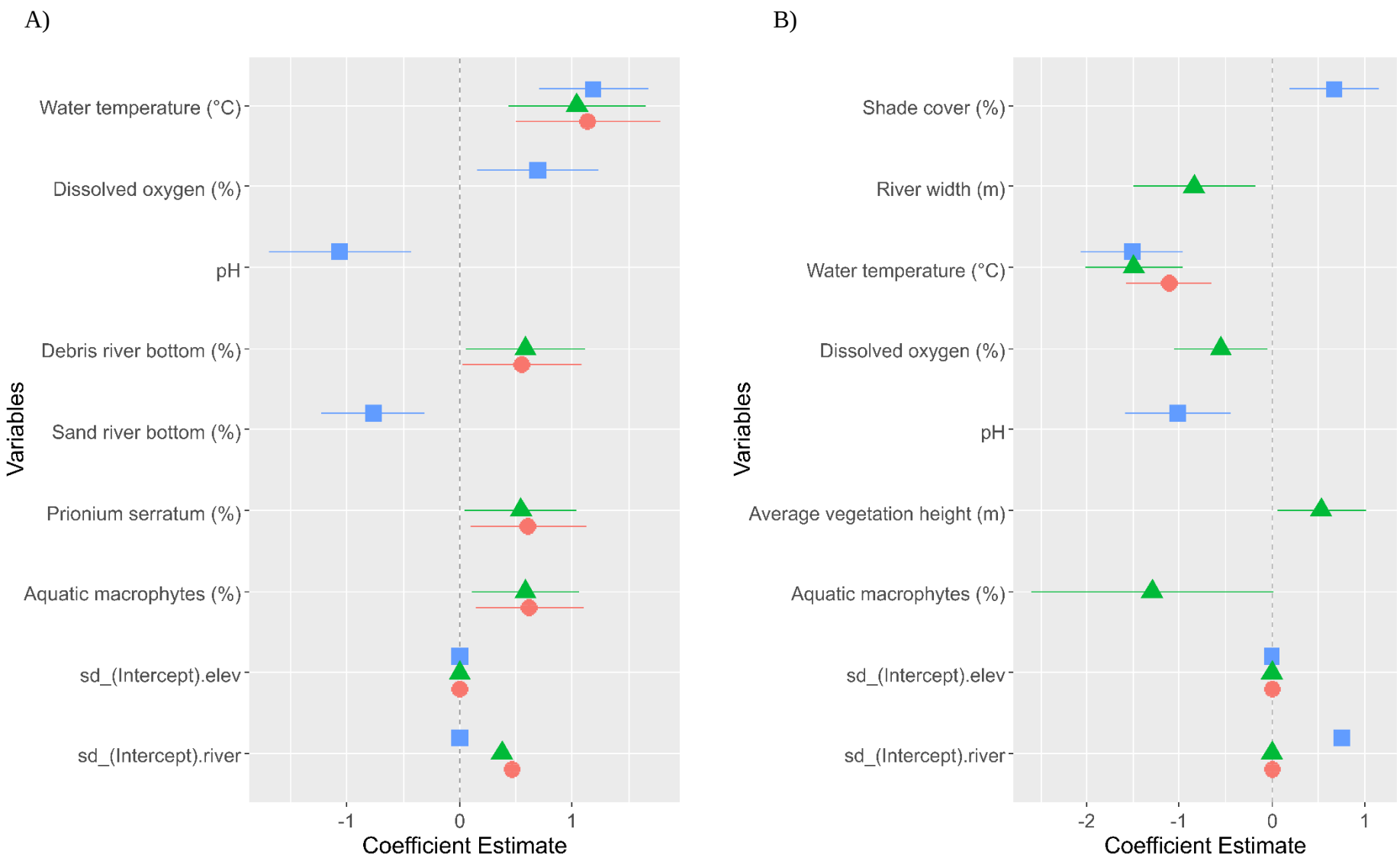

Fig. 2.7 Significant environmental variables influencing A) Shannon index scores and B) DBI scores.

Squares = larvae; triangles = adults; circle = adjusted adults.

Table 2.3 Assemblage analyses using multivariate generalised linear modelling of the effects of environmental variables on dragonfly larvae, adults, and adjusted adults. Bolded numbers signify significance.

	Deviation test statistic		
Variables	**Larvae**	**Adults**	**Adjusted adults**
Flow type	**109.49*** **	**145.7*** **	**134.24*** **
% Shade	**87.37*** **	**75.20*** **	**73.17*** **
Width	**79.08*** **	**69.57*** **	**67.03*** **
Depth			
Water measures			
Water temperature	**234.52*** **	**258.80*** **	**315.15*** **
Dissolved oxygen	**70.85*** **		**80.92** **
Conductivity			
pH	**137.96*** **	**62.93*** **	**57.27** **
River edge cover			
% Debris			
% Sand		**43.89***	
% Rock	**71.52*** **	**80.00*** **	**65.87** **
River bed cover			
% Debris	**43.42*** **		
% Sand	**44.46** **		
% Rock	**137.67*** **	**95.78*** **	**77.64*** **
Vegetation			
Avge veg height			
Prionium serratum		**104.74*** **	**153.43*** **
Aquatic macrophytes		**97.46*** **	**110.53*** **

* P < 0.05, ** P < 0.01, *** P < 0.001

2.4 Discussion

2.4.1 'Father knows best' hypothesis

For rivers in the Cape Floristic Region (CFR), larvae and adult dragonfly assemblages followed the same trajectory, and positively correlated for abundance, species richness, Shannon diversity, and DBI scores. Despite this, when the site scale was too small (here 10 x 3 m) these relationships were weak, especially for DBI scores. At the large scale (here 450 x 3 m), correlations were very strong, and although significant at the 5% level for abundance and richness, they were not significant for Shannon diversity and DBI. Moderately strong, significant correlations occurred at the medium scale (90 x 3 m). These differences reiterate the importance of considering sampling scales, especially for highly mobile insects such as dragonflies. Regardless of spatial scale, the relationship was always positive for assemblage parameters between larvae and adults. This could be due to both assemblages having the same main environmental drivers (water temperature, which positively correlated with agricultural land transformation). Abundance and species richness of dragonflies increased with river size (width and depth). This is in accordance with the biogeographical principle, that a larger area supports more species, which is well established for freshwater ecosystems, including rivers (Lassen, 1975; Brönmark et al., 1984; Brönmark, 1985; Hugueny, 1989; Rey Benayas et al., 1999) and even for dragonflies specifically (Oertli et al., 2002; Chovanec et al., 2004). Furthermore, the 'habitat-heterogeneity hypothesis' suggests that a variety of different biotopes and features will lead to high species diversity (Tews et al., 2004; Cramer & Willig, 2005). Sites with many microhabitats, in terms of both aquatic and terrestrial variables, would fulfil the requirements for a wide range of dragonfly species.

At all three scales, the 'father knows best' hypothesis is disproved. Where a species was observed at a site, whether as a larva or an adult, there was never a significantly greater chance of the same species, but opposite life stage, occurring at the same site. Despite this, the proportion of matches did increase with spatial scale. The unavoidable differences in sampling techniques and effort make comparisons between larvae and adults difficult (Hawking & New, 1999). The mobile, conspicuous adults are difficult to miss during waterside surveys that cover the entire site area, whereas the well-camouflaged larvae are more difficult to sample from their in-water habitats, and where only a subset of the site is usually sampled (Simaika & Samways, 2012; Samways & Simaika, 2016). This is made more difficult by the extremely

specialised requirements of some species. Species like *Trithemis dorsalis/furva* and *Platycypha fitzimonsi* were more strongly represented in our adult surveys than in larvae surveys, whereas others like *Zygonyx natalensis* and *Paragomphus cognatus* appeared mostly in larval samples. This could be related to behaviour such as differences in habitat requirements, territory guarding or perching, egg-laying habits or the degree to which adults utilise the rivers for mate finding, foraging, and ovipositing (Luke et al., 2017). For example, some Gomphidae species rarely visit rivers except to oviposit, and spend much of their aerial phase foraging elsewhere across the landscape, making adult records rarer (Orr, 2006), as seen here for *P. cognatus* and *Ceratogomphus pictus*. Furthermore, the highly mobile adults disperse freely, and it is possible that some of the recorded individuals were feeding or just passing through sub-optimal breeding sites, whereas presence of larvae guarantees they are developing in the area (Valente-Neto et al., 2016). Alternatively, perhaps weak competing adults were present at sub-optimal sites because they were driven out from more optimal sites by more competitive individuals. Therefore, females may not be looking to mate when feeding, or choose not to mate with the males who have sub-optimal territories, and so larvae may have been absent from these sites. Only 118 exuviae from five species were sampled here. Bried et al. (2012) concluded that lower species detection probability for exuviae would typically result in species richness observations that are biased low. It is not entirely clear why exuviae are so scarce in the CFR because in many other parts of the world (such as Europe) they are far more abundant (Raebel et al., 2010). Perhaps this is due to a combination of factors, such as fluctuating water levels, strong water flows, scavengers, our lack of knowledge of their behaviour and/or selecting cryptic locations at the time of adult emergence.

The different approaches to biodiversity surveys of the dragonflies attempted here were not interchangeable, as conveyed by the mismatching between larvae and adult species at the small spatial scales. Furthermore, DBI scores were weakly correlated at these small scales between larvae and adults, and between larvae and adjusted adults. The DBI is highly sensitive as it uses the species level, and exclusion of a single species may therefore have a large effect on the value of the site score. Although species estimates and rarefaction curves showed sampling effort to be adequate for both larvae and adults, there were still six dragonfly species observed as adults but not as larvae. Of these, *Z. minuscula* and *C. umbratus* both have a high DBI score and their absence in larval samples would greatly alter the site scores (Samways & Simaika, 2016). However, absence of larvae for these six taxa may be due to the taxonomic challenge, with some South African larvae not yet described (Clark & Samways, 1996). Furthermore,

larvae are difficult to identify to species level, which could result in misidentification. Usually only last instar larvae can be used, and the numerous discarded individuals that were too small to identify may have influenced the results. Alternatively, differences between larvae and adult scores at a site could be the result of actual differences in the species assemblages. This could be due to many factors, including the dispersal ability of adults. Adult dragonflies are highly mobile and although species like *P. flavescens* were recorded in the rivers here as adults, they have larvae that generally develop in temporary pools and not in rivers (Johansson & Suhling, 2004).

2.4.2 'Shared environmental preferences' hypothesis

Despite species differences at sites, water temperature was positively correlated with agricultural land use, and these were the most influential, being the only variables that significantly influenced all the diversity measures (abundance, species richness, Shannon and DBI scores) and the assemblages of both larvae and adults. Here, abundance, species richness, and diversity positively correlated with water temperature and agricultural land use. Warmer water temperatures are associated with higher productivity and promote faster egg development (Pritchard & Leggott, 1987), as well as faster growth of larvae (Suhling et al., 2015). Net thermal radiation is a key determinant of water temperature (Karr & Schlosser, 1978). Adult dragonflies, especially those that thermoregulate exogenously, are sensitive to the thermal landscape, and select suitable microhabitats accordingly (May, 1978).

The great differences in microhabitats experienced along CFR rivers cause in-water conditions in the lower reaches to vary from those of the mountainous upper reaches. Lower reaches characteristically have warmer water temperatures due to lower elevations, wider, less turbulent sections, and less shading from trees (Dallas & Day, 2007). These sections are often largely transformed, and agricultural development is likely to increase water temperature due to reduced riparian shade, with shade being an important driver of dragonfly assemblages (Remsburg et al., 2008; Piggott et al., 2012). Higher species richness, abundance and diversity of larvae and adults occurred at agriculturally transformed areas compared to natural sites. These river sections were larger than the natural sections with more habitat space to occupy. Species that are sensitive to disturbance and temperature were absent and replaced by the habitat generalist species that dominated the agricultural areas (Júnior et al., 2015). Furthermore, larval and adult DBI scores negatively correlated with water temperature and agricultural land transformation. The DBI is a measure of habitat quality, with the lower scores

in the transformed areas resulting from the absence of sensitive, habitat specialists such as *P. fitzimonsi* and *C. conspicuus*. The in-water parameters of pH and dissolved oxygen were important drivers for both larvae and adult assemblages, which can also be directly linked to water quality.

Dragonfly larvae and adults only shared a few of the same important variables (water temperature, flow type, shade cover, river width, pH, and percentage rocks on the side of the river and on the riverbed), meaning the 'shared environmental preferences' hypothesis is rejected. Other than the shared variables between both stages, adult assemblages were influenced greatly by vegetation-related characteristics. Taller vegetation decreased adult abundance but increased DBI scores, possibly related to shading and water temperature effects (Niba & Samways, 2006; Rivers-Moore et al., 2018), with the presence of taller vegetation (such as *Brabejum stellatifolium*) in the natural, untransformed sections inhabited by endemic, habitat specialists (Meek et al., 2013). The indigenous Palmiet reed, *P. serratum*, plays an important functional role in CFR river systems (Kietzka et al., 2017) and was key driver of community composition, abundance, species richness and diversity of adult dragonflies. It is favoured as a perching site, and is particularly important for some endemic species such as *P. furcigerum* (Rambur, 1842) (Samways & Sharratt, 2010). The presence of other aquatic vegetation was also important for adults as both perching, but more importantly, as suitable oviposition sites (Corbet, 1999; Clausnitzer et al., 2009; Luke et al., 2017). Vegetation-related variables were not vital drivers of the aquatic larvae but river bed substrates were, as shown in other studies (Huggins & DuBois, 1982). These differences may indicate that habitat selection is based on the microhabitat features that are important for that specific life stage. In other words, habitat selection by adults could be based on features that are important for the adult stage, suggesting that they may not necessarily be looking for suitable sites for their offspring. Similarly, after larvae have hatched they may disperse to nearby areas with favourable substrate types.

2.4.3 Significance of using the Dragonfly Biotic Index

The DBI is based on presence vs. absence of the particular suite of dragonfly species at localities being compared spatially, and temporally in the case of restoration. While the DBI is based primarily on use of adults (mostly male, but also female) for practical and logistical purposes, it does not exclude the gathering of supplementary data on larvae and/or exuviae, as

explained in the manual for its use (Samways & Simaika, 2016). The importance of gathering these extra data is emphasized here with some extra species recorded as larvae in sites where the adults were not found, notwithstanding the challenges of gathering juvenile-stage data. Furthermore, hawking, cruising, and canopy resting or foraging adults in the vicinity of the sampling site should also be recorded.

2.5 Conclusion

I have shown that for dragonflies in rivers of the CFR, larvae and adult surveys were not always interchangeable. The interchangeability of larvae and adult surveys is dependent on the type of question being asked, and the appropriateness of the scale being used. Dragonfly larvae and adults correlated for abundance, species richness, Shannon diversity, and DBI scores, which strengthened as spatial scale increased. Furthermore, both life stages were driven by water temperature, which positively correlated with agricultural land use. This reiterates their use, at any life stage, as good indicators of anthropogenic land transformation. These trends suggest that for coarser ecological questions, where sampling at larger scales is appropriate, either the larvae or adults can be used. For example, to determine whether dragonfly species richness differs between natural and agricultural reaches, surveys could be conducted at larger scales, and can use one or the other life stage. The preferred life stage would be dependent on factors such as sampling season, favoured sampling method, river accessibility, and degree of expertise in identifying the different life stages. Despite this, particularly at smaller scales, larvae and adult species differed in site occurrences and in their environmental drivers. Therefore, for complete taxonomic assessments or for fine scale ecological questions, it is important to survey both life stages. Overall, these results bear out the DBI recommendations, especially in view of its aim being principally to record *all* dragonfly species present at designated site, and to include all life stages if possible, microhabitats, and species locations both at the water and away from it.

2.6 References

Allan, J. D., & Flecker, A. S. (1993). Biodiversity conservation in running waters. BioScience, 43(1), 32-43.

Agouridis, C. T., Wesley, E. T., Sanderson, T. M., & Newton, B. L. (2015). Aquatic Macroinvertebrates: Biological Indicators of Stream Health. University of Kentucky College of Agriculture, Lexington, KY, USA.

Bates, D., Maechler, M., Bolker, B., & Walker, S. (2014). lme4: Linear mixed-effects models using Eigen and S4. R package version, 1(7), 1-23.

Bates, D., Sarkar, D., Bates, M. D., & Matrix, L. (2007). The lme4 package. R package version, 2(1), 74.

Bird, M. S., Mlambo, M. C., Wasserman, R. J., Dalu, T., Holland, A. J., Day, J. A., Villet, M. H., Bilton, D. T., Barber-James, H. M., & Brendonck, L. (2019). Deeper knowledge of shallow waters: reviewing the invertebrate fauna of southern African temporary wetlands. Hydrobiologia, 827(1), 89-121.

Bolker, B. M., Brooks, M. E., Clark, C. J., Geange, S. W., Poulsen, J. R., Stevens, M. H. H., & White, J. S. S. (2009). Generalized linear mixed models: a practical guide for ecology and evolution. Trends in Ecology and Evolution, 24(3), 127-135.

Bried, J. T., D'amico, F., & Samways, M. J. (2012). A critique of the dragonfly delusion hypothesis: why sampling exuviae does not avoid bias. Insect Conservation and Diversity, 5(5), 398-402.

Briggs, A. J., Pryke, J. S., Samways, M. J., & Conlong, D. E. (2019). Complementarity among dragonflies across a pondscape in a rural landscape mosaic. Insect Conservation and Diversity.

Brito, J. G., Martins, R. T., Oliveira, V. C., Hamada, N., Nessimian, J. L., Hughes, R. M., Ferraz, S. F., & de Paula, F. R. (2018). Biological indicators of diversity in tropical streams: Congruence in the similarity of invertebrate assemblages. Ecological Indicators, 85, 85-92.

Brönmark, C. (1985). Freshwater snail diversity: effects of pond area, habitat heterogeneity and isolation. Oecologia, 67(1), 127-131.

Brönmark, C., Herrmann, J., Malmqvist, B., Otto, C., & Sjöström, P. (1984). Animal community structure as a function of stream size. Hydrobiologia, 112(1), 73-79.

Chovanec, A., Waringer, J., Raab, R., & Laister, G. (2004). Lateral connectivity of a fragmented large river system: assessment on a macroscale by dragonfly surveys

(Insecta: Odonata). Aquatic Conservation: Marine and Freshwater Ecosystems, 14(2), 163-178.

Clark, T. E., & Samways, M. J. (1996). Dragonflies (Odonata) as indicators of biotope quality in the Kruger National Park, South Africa. Journal of applied ecology, 1001-1012.

Clausnitzer, V. (2003). Dragonfly communities in coastal habitats of Kenya: indication of biotope quality and the need of conservation measures. Biodiversity and Conservation, 12(2), 333-356.

Clausnitzer, V., Kalkman, V. J., Ram, M., Collen, B., Baillie, J. E., Bedjanič, M., Darwall, W.R., Dijkstra, K.D.B., Dow, R., Hawking, J., & Karube, H. (2009). Odonata enter the biodiversity crisis debate: the first global assessment of an insect group. Biological Conservation, 142(8), 1864-1869.

Clausnitzer, V., Simaika, J. P., Samways, M. J., & Daniel, B. A. (2017). Dragonflies as flagships for sustainable use of water resources in environmental education. Applied Environmental Education and Communication, 16, 196-209.

Conrad, K. F., Willson, K. H., Harvey, I. F., Thomas, C. J., & Sherratt, T. N. (1999). Dispersal characteristics of seven odonate species in an agricultural landscape. Ecography, 22(5), 524-531.

Corbet, P. S. (1999). Dragonflies: Behavior and Ecology of Odonata. Cornell University press, Ithaca, NY.

Cramer, M. J., & Willig, M. R. (2005). Habitat heterogeneity, species diversity and null models. Oikos, 108(2), 209-218.

Dallas, H. F. & Day, J. A. (2007) Natural variation in macroinvertebrate assemblages and the development of a biological banding system for interpreting bioassessment data – a preliminary evaluation using data from upland sites in the south-western Cape, South Africa. Hydrobiologia, 575, 231-244.

de Moor, F. C. (2017). Dragonflies as indicators of aquatic ecosystem health. South African Journal of Science, 113, 1-2.

Dickens C. W. S, & Graham, P. M. (1998) Biomonitoring for effective management of wastewater discharges and the health of the river environment. Aquatic Ecosystem Health and Management, 1, 199-217.

Dickens, C. W. S., & Graham, P. M. (2002). The South African Scoring System (SASS) version 5 rapid bioassessment method for rivers. African Journal of Aquatic Science, 27, 1-10.

Dunkle, S. W. (1976). Larva of the dragonfly, *Ophiogomphus arizonicus* (Odonata: Gomphidae). Florida Entomologist, 59, 317-320.

Harabiš, F., & Dolný, A. (2012). Human altered ecosystems: suitable habitats as well as ecological traps for dragonflies (Odonata): the matter of scale. Journal of Insect Conservation, 16(1), 121-130.

Hardersen, S. (2008). Dragonfly (Odonata) communities at three lotic sites with different hydrological characteristics. Italian Journal of Zoology, 75, 271-283.

Hawking, J. H., & New, T. R. (1999). The distribution patterns of dragonflies (Insecta: Odonata) along the Kiewa River, Australia, and their relevance in conservation assessment. Hydrobiologia, 392(2), 249-260.

Hortal, J., Borges, P. A. V., & Gaspar, C. (2006). Evaluating the performance of species richness estimators: sensitivity to sample grain size. Journal of Animal Ecology, 75, 274-287.

Huggins, D. G., & DuBois, M. D. (1982). Factors affecting microdistribution of two species of burrowing dragonfly larvae, with notes on their biology (Anisoptera: Gomphidae). Odonatologica, 11(1), 1-14.

Hugueny, B. (1989). West African rivers as biogeographic islands: species richness of fish communities. Oecologia, 79(2), 236-243.

Johansson, F., & Suhling, F. (2004). Behaviour and growth of dragonfly larvae along a permanent to temporary water habitat gradient. Ecological Entomology, 29(2), 196-202.

Júnior, P. D. M., Batista, J. D., & Cabette, H. S. R. (2015). Community assembly of adult odonates in tropical streams: an ecophysiological hypothesis. PloS One, 10(4), e0123023.

Kalkman, V. J., Boudot, J. P., Bernard, R., De Knijf, G., Suhling, F., & Termaat, T. (2018). Diversity and conservation of European dragonflies and damselflies (Odonata). Hydrobiologia, 811(1), 269-282.

Karr, J. R., & Schlosser, I. J. (1978). Water resources and the land-water interface. Science, 201(4352), 229-234.

Keetsch, D. P., & Moran, V. C. (1966). Observations on the biology of nymphs of Paragophus cognatus (Rambur) (Odonata: Gomphidae). I. Habitat selection in relation to substrate particle size. Proceedings of the Royal Entomological Society of London, 41, 116-122.

Kemp, D. J. (2018). Habitat selection and territoriality. In insect behavior: from mechanisms to ecological and evolutionary consequences. Oxford University Press (pp. 80 - 93), Oxford, UK.

Kietzka, G. J., Pryke, J. S., & Samways, M. J. (2017). Aerial adult dragonflies are highly sensitive to in-water conditions across an ancient landscape. Diversity and Distributions, 23(1), 14-26.

Kietzka, G. J., Pryke, J. S., & Samways, M. J. (2018). Comparative effects of urban and agricultural land transformation on Odonata assemblages in a biodiversity hotspot. Basic and Applied Ecology, 33, 89-98.

Lassen, H. H. (1975). The diversity of freshwater snails in view of the equilibrium theory of island biogeography. Oecologia, 19(1), 1-8.

Lemelin, R. H. (2007). Finding beauty in the dragon: the role of dragonflies in recreation and tourism. Journal of Ecotourism, 6(2), 139-145.

Lesch, V., & Bouwman, H. (2018). Adult dragonflies are indicators of environmental metallic elements. Chemosphere, 209, 654-665.

Lubertazzi, M. A. A., & Ginsberg, H. S. (2009). Persistence of dragonfly exuviae on vegetation and rock substrates. Northeastern Naturalist, 16(1), 141 – 148.

Luke, S. H., Dow, R. A., Butler, S., Vun Khen, C., Aldridge, D. C., Foster, W. A., & Turner, E. C. (2017). The impacts of habitat disturbance on adult and larval dragonflies (Odonata) in rainforest streams in Sabah, Malaysian Borneo. Freshwater Biology, 62(3), 491-506.

May, M. L. (1978). Thermal adaptations of dragonflies. Odonatologica, 7(1), 27-47.

Maynou, X., Martín, R., & Aranda, D. (2017). The role of small secondary biotopes in a highly fragmented landscape as habitat and connectivity providers for dragonflies (Insecta: Odonata). Journal of Insect Conservation, 21(3), 517-530.

Mazerolle, M. J., & Mazerolle, M. M. J. (2017). Package 'AICcmodavg'. R package.

McCauley, S. J., Hammond, J. I., & Mabry, K. E. (2018). Simulated climate change increases larval mortality, alters phenology, and affects flight morphology of a dragonfly. Ecosphere, 9(3), e02151.

Meek, C. S., Richardson, D. M., & Mucina, L. (2013). Plant communities along the Eerste River, Western Cape, South Africa: Community descriptions and implications for restoration. Koedoe, 55(1), 01-14.

Morrison III, W. R., Waller, J. T., Brayshaw, A. C., Hyman, D. A., Johnson, M. R., & Fraser, A. M. (2018). Evaluating multiple arthropod taxa as indicators of invertebrate diversity in old fields. The Great Lakes Entomologist, 45(1), 56-68.

Niba, A. S., & Samways, M. J. (2006). Remarkable elevational tolerance in an African Odonata larval assemblage. Odonatologica, 35(3), 265-280.

Oertli, B., Joye, D. A., Castella, E., Juge, R., Cambin, D., & Lachavanne, J. B. (2002). Does size matter? The relationship between pond area and biodiversity. Biological Conservation, 104(1), 59-70.

Oksanen, J., Blanchet, F. G., Kindt, R., Legendre, P., Minchin, P. R., O'hara, R. B., Simpson, G. L., Solymos, P., Stevens, M. H. H., Wagner, H., & Oksanen, M. J. (2015). Package 'vegan'. Community ecology package, version, 2(9).

Orr, A. G. (2006). Odonata in Bornean tropical rain forest formations: diversity, endemicity and implications for conservation management. In forest and dragonflies. Pensoft Publishers (pp. 51-78), Sofia, Bulgaria.

Osborn, R., & Samways, M. J. (1996). Determinants of adult dragonfly assemblage patterns at new ponds in South Africa. Odonatologica, 25(1), 49-58.

Patten, M. A., Bried, J. T., & Smith-Patten, B. D. (2015). Survey data matter: predicted niche of adult vs breeding Odonata. Freshwater Science, 34(3), 1114 – 1122.

Piggott, J. J., Lange, K., Townsend, C. R., & Matthaei, C. D. (2012). Multiple stressors in agricultural streams: a mesocosm study of interactions among raised water temperature, sediment addition and nutrient enrichment. PloS One, 7(11), e49873.

Pritchard, G., & Leggott, M. A. (1987). Temperature, incubation rates and origins of dragonflies. Advances in Odonatology, 3(1), 121-126.

QGIS Development Team. (2014). QGIS Geographic Information System. Open Source Geospatial Foundation Project.

R Core Team. (2018). R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria.

Raebel, E. M., Merckx, T., Riordan, P., Macdonald, D. W., & Thompson, D. J. (2010). The dragonfly delusion: why it is essential to sample exuviae to avoid biased surveys. Journal of Insect Conservation, 14(5), 523-533.

Remsburg, A. J., Olson, A. C., & Samways, M. J. (2008). Shade alone reduces adult dragonfly (Odonata: Libellulidae) abundance. Journal of Insect Behavior, 21(6), 460-468.

Renner, S., Sahlén, G., & Périco, E. (2016). Testing dragonflies as species richness indicators in a fragmented subtropical Atlantic forest environment. Neotropical Entomology, 45(3), 231-239.

Rey Benayas, J. M., Colomer, M. G., & Levassor, C. (1999). Effects of area, environmental status and environmental variation on species richness per unit area in Mediterranean wetlands. Journal of Vegetation Science, 10(2), 275-280.

Rivers-Moore, N. A., Dallas, H. F., de Moor, F. C., & Barendse, J. (2018). Relationships of water temperature and aquatic macroinvertebrate community structure with non-native riparian plant densities in the southern Cape, South Africa. African Journal of Aquatic Science, 43(3), 215-227.

Rosset, V., Simaika, J. P., Arthaud, F., Bornette, G., Vallod, D., Samways, M. J., & Oertli, B. (2013). Comparative assessment of scoring methods to evaluate the conservation value of pond and small lake biodiversity. Aquatic Conservation: Marine and Freshwater Ecosystems, 23, 23-36.

Samways, M. J. (1989). Farm dams as nature reserves for dragonflies (Odonata) at various altitudes in the Natal Drakensberg Mountains, South Africa. Biological Conservation, 48, 181-187.

Samways, M. J. (1992). Dragonfly conservation in South Africa: a biogeographical perspective. Odonatologica, 21(2), 165-180.

Samways, M. J. (2008). Dragonflies and damselflies of South Africa (No. 70). Pensoft Publishers, Sophia, Bulgaria.

Samways, M. J., & Sharratt, N. J. (2010). Recovery of endemic dragonflies after removal of invasive alien trees. Conservation Biology, 24(1), 267-277.

Samways, M. J., & Simaika, J. P. (2016). Manual of freshwater assessment for South Africa: Dragonfly biotic index. South African National Biodiversity Institute, Pretoria, South Africa.

Samways, M. J., & Steytler, N. S. (1996). Dragonfly (Odonata) distribution patterns in urban and forest landscapes, and recommendations for riparian management. Biological Conservation, 78, 279-288.

Samways, M. J., & Taylor, S. (2004). Impacts of invasive alien plants on Red-Listed South African dragonflies (Odonata): working for water. South African Journal of Science, 100(1-2), 78-80.

Shafie, M. S., Wong, A. B., Harun, S., & Fikri, A. H. (2017). The use of aquatic insects as bio–indicator to monitor freshwater stream health of Liwagu River, Sabah, Malaysia. Journal of Entomology and Zoology Studies, 5(4), 1662-1666.

Simaika, J. P., & Samways, M. J. (2009a). An easy-to-use index of ecological integrity for prioritizing freshwater sites and for assessing habitat quality. Biodiversity and Conservation, 18(5), 1171-1185.

Simaika, J. P., & Samways, M. J. (2009b). Reserve selection using Red Listed taxa in three global biodiversity hotspots: dragonflies in South Africa. Biological Conservation, 142(3), 638-651.

Simaika, J. P., & Samways, M. J. (2011). Comparative assessment of indices of freshwater habitat conditions using different invertebrate taxon sets. Ecological Indicators, 11(2), 370-378.

Simaika, J. P., & Samways, M. J. (2012). Using dragonflies to monitor and prioritize lotic systems: a South African perspective. Organisms, Diversity & Evolution, 12(3), 251-259.

Simaika, J. P., Samways, M. J., & Frenzel, P. P. (2016). Artificial ponds increase local dragonfly diversity in a global biodiversity hotspot. Biodiversity and Conservation, 25(10), 1921-1935.

Smith, J., Samways, M. J., & Taylor, S. (2007). Assessing riparian quality using two complementary sets of bioindicators. Biological Conservation, 16, 2695-2713.

Stewart, D. A., & Samways, M. J. (1998). Conserving dragonfly (Odonata) assemblages relative to river dynamics in an African savanna game reserve. Conservation Biology, 12(3), 683-692.

Steytler, N. S., & Samways, M. J. (1995). Biotope selection by adult male dragonflies (Odonata) at an artificial lake created for insect conservation in South Africa. Biological Conservation, 72(3), 381-386.

Stoks, R., & Cordoba-Aguilar, A. (2012). Evolutionary ecology of Odonata: a complex life cycle perspective. Annual Review of Entomology, 57, 249-265.

Suh, A. N., & Samways, M. J. (2001). Development of a dragonfly awareness trail in an African botanical garden. Biological Conservation, 100(3), 345-353.

Suhling, F., Müller, O., & Martens, A. (2014). The dragonfly larvae of Namibia (Odonata). Libellula Supplement, 13, 5-106.

Suhling, F., Suhling, I., & Richter, O. (2015). Temperature response of growth of larval dragonflies–an overview. International Journal of Odonatology, 18(1), 15-30.

ter Braak, C. J. (1990). Update notes: CANOCO, version 3.10. Agricultural Mathematics Group, Wageningen, Holland, 35.

ter Braak, C. J. F., & Šmilauer, P. (2012). Canoco 5, Windows release (5.00). Software for mutivariate data exploration, testing, and summarization. Biometris, Plant Research International, Wageningen.

Tews, J., Brose, U., Grimm, V., Tielbörger, K., Wichmann, M. C., Schwager, M., & Jeltsch, F. (2004). Animal species diversity driven by habitat heterogeneity/diversity: the importance of keystone structures. Journal of Biogeography, 31(1), 79-92.

Uys, M. U., Goetsch, M. A., & O'Keeffe, J. H. (1996). Ecological Indicators, a Review and Recommendations. National Biomonitoring Programme for Riverine Ecosystems. Report Series No. 4. Institute for Water Quality Studies, Department of Water Affairs and Forestry. Pretoria, South Africa, 93.

Valente-Neto, F., de Oliveira Roque, F., Rodrigues, M. E., Juen, L., & Swan, C. M. (2016). Toward a practical use of Neotropical odonates as bioindicators: Testing congruence across taxonomic resolution and life stages. Ecological Indicators, 61, 952-959.

Vavrek, M. J. (2011). Fossil: palaeoecological and palaeogeographical analysis tools. Palaeontologia Electronica, 14.

Wang, Y. I., Naumann, U., Wright, S. T., & Warton, D. I. (2012). Mvabund – an R package for model-based analysis of multivariate abundance data. Methods in Ecology and Evolution, 3(3), 471-474.

Wickham, H., & Chang, W. (2008). ggplot2: An implementation of the grammar of graphics. R package version 0.7.

Wishart, M. J., & Day, J. A. (2002). Endemism in the freshwater fauna of the south–western Cape, South Africa. Limnologie, 28(4), 1–5.

2.7 Appendices

Appendix 2.A Correlations between larvae (Y axis) and adults (X axis) at three spatial scales.

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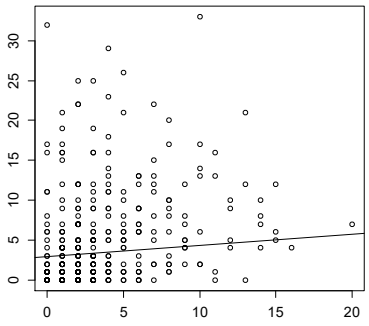
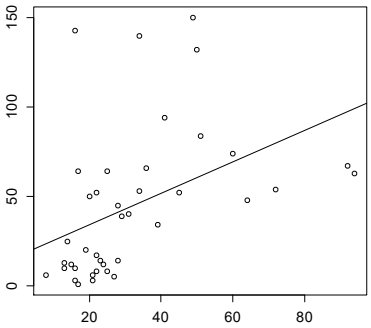
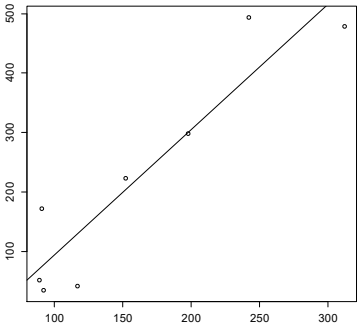
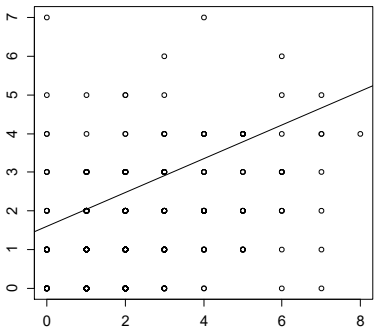
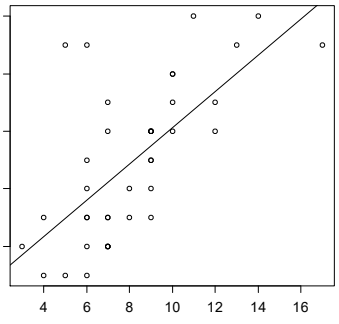
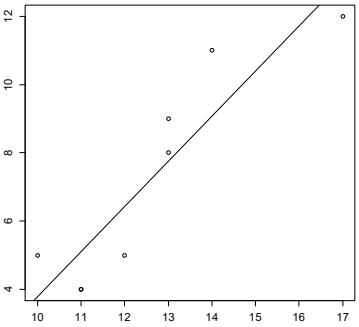
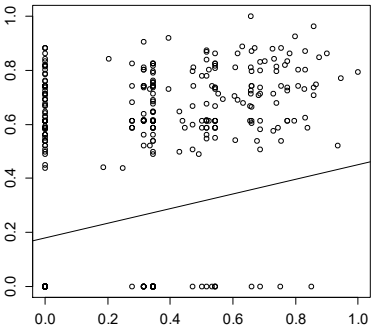
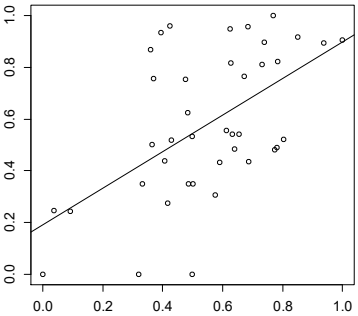
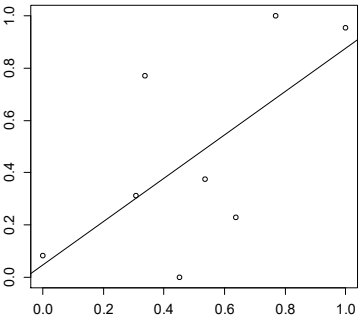
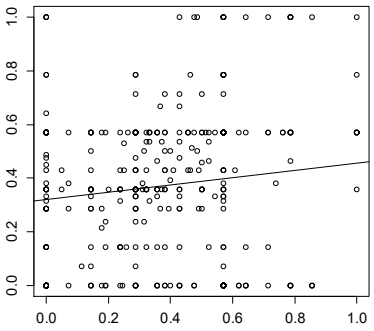
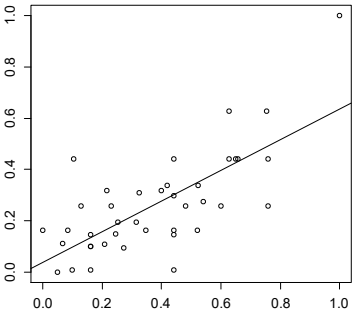
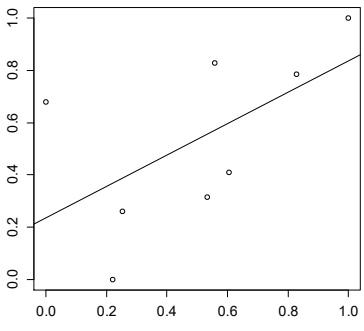
Abundance			
Species richness			
Shannon index			
DBI			

Appendix 2.B Correlations between larvae (Y axis) and adjusted adults (X axis) at three spatial scales.

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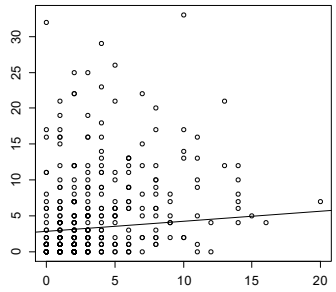
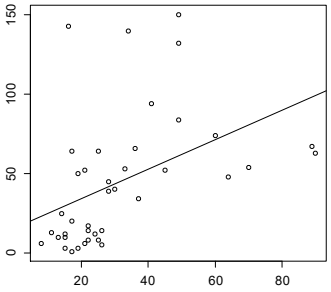
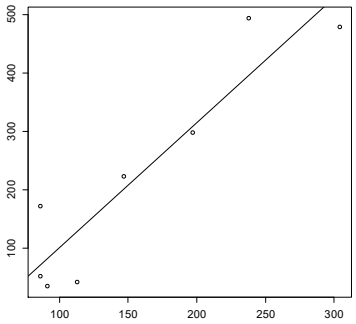
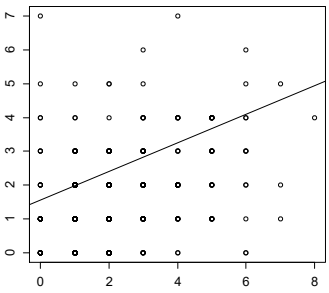
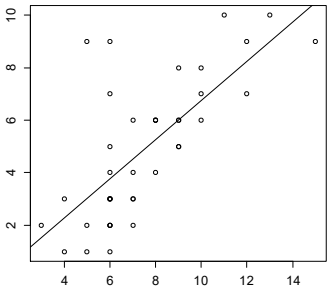
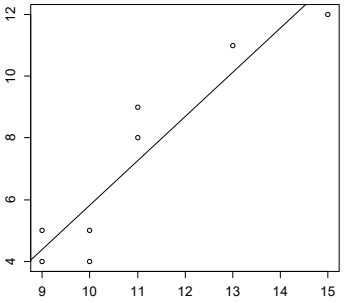
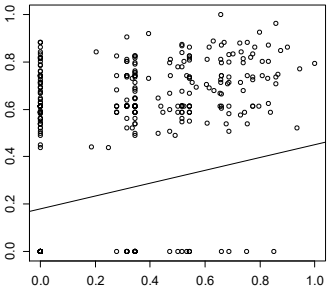
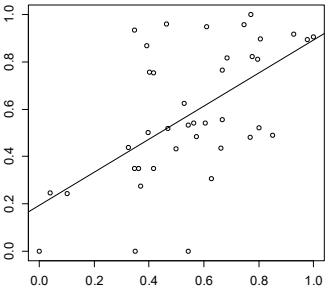
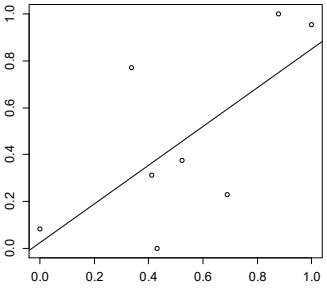
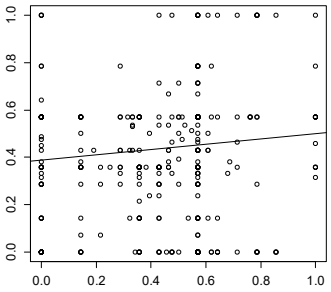
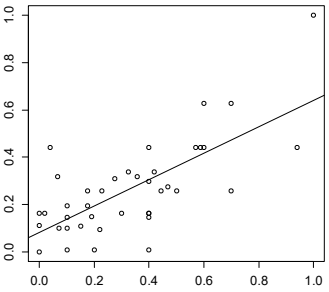
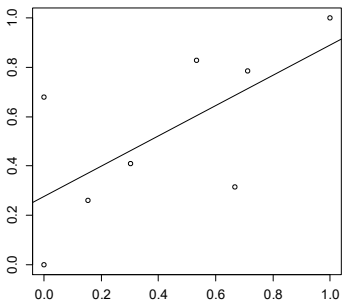
Abundance			
Species richness			
Shannon index			
DBI			

Appendix 2.C Species names and codes.

Scientific name	Species code
Suborder ZYGOPTERA	
Family CHLOROCYPHIDAE	
Platycypha fitzsimonsi (Pinhey 1950)	Pfi
Family SYNLESTIDAE	
Chlorolestes conspicuous (Hage in Selys 1862)	CC
Chlorolestes umbratus (Selys 1862)	CU
Family PROTONURIDAE	
Elattoneura glauca (Selys 1862)	EG
Elattoneura frenulata (Hage in Selys 1862)	EF
Family PLATYCNEMIDIDAE	
Allocnemis leucosticta (Selys 1863)	AL
Family COENAGRIONIDAE	
Ceriagrion glabrum (Burmeister 1839)	CG
Pseudagrion draconis (Barnard 1937)	PD
Pseudagrion furcigerum (Rambur 1842)	PF
Pseudagrion kerteni (Gersaecker 1869)	PK
Suborder ANISOPTERA	
Family AESHNIDAE	
Zosteraeschna minuscula (McLachlan 1896)	ZM
Pinheyschna subpupillata (McLachlan 1896)	PS
Anax imperator (Rambur 1842)	AI
Anax speratus (Hagen 1867)	AS
Family GOMPHIDAE	
Ceratogomphus pictus (Selys 1854)	CP
Paragomphus cognatus (Rambur 1842)	PC
Family CORDULIIDAE	
Syncordulia gracilis (Burmeister 1839)	SG
Family LIBELLULIDAE	
Orthetrum julia capicola (Kimmins 1957)	OJ
Crocothemis erythraea (Brullé 1832)	CE
Crocothemis sanguinolenta (Burmeister 1839)	CS
Trithemis arteriosa (Burmeister 1839)	TA
Trithemis dorsalis/furva (Rambur 1842/Karsch 1899)	TD
Zygonyx natalensis (Martin 1900)	ZN
Pantala flavescens (Fabricius 1798)	Pfl
Tramea limbata (Desjardins 1832)	TL

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Chapter 3: Applying the umbrella index across aquatic insect taxon sets for freshwater assessment

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Abstract

Biological surrogates in conservation biology are valuable for rapid biodiversity and environmental surveys, and as an early warning of potential threats. However, these surrogates need to be simple and inexpensive to apply. The umbrella index was applied here to quantify the selection of surrogate species for biodiversity assessments, but required interrogation for application in areas rich in threatened endemic species. Aquatic larvae of Ephemeroptera, Plecoptera and Trichoptera (EPT), as well as the adult Odonata, are all highly responsive to changes in freshwater condition. Using the umbrella index, we evaluated the performance of the surrogate species approach for aquatic insect conservation in a region (Greater Cape Floristic Region) with an exceptional level of rare and endemic species, across multiple rivers with different disturbance levels. Due to a lack of species level information, EPT taxa scores were calculated using morphospecies within families, and Odonata were identified to species level. The umbrella index identified eight EPT species and seven Odonata species as potential surrogates. Both these groups co-occurred with high percentages of their own overall groups (EPT surrogates for overall EPT, and Odonata surrogates for overall Odonata), as well as for the other group (EPT surrogates for overall Odonata, and Odonata surrogates for overall EPT). The index was surprisingly flexible, and performed well in an area with so many species of conservation concern, as well as across spatial scales greater than a single river, with varying degrees of disturbance. Both EPT taxa and Odonata showed promise as potential biodiversity surrogates, and for future conservation planning. Ideally, conservationists should aim to use taxa that are easy to identify to species level, with known sensitivities to human disturbance. However, when this is not a possibility, the umbrella index is still applicable and accurate for morphospecies.

Key words: biodiversity hotspot; habitat transformation; Odonata; Ephemeroptera; Plecoptera, Trichoptera

3.1 Introduction

Freshwater ecosystems play vital roles, both ecologically, and for the services they provide to humans (Pringle et al., 2000; Russi et al., 2013; Harrison et al., 2016). Rivers are our main source of renewable freshwater, and are among the most threatened ecosystems (Pringle et al., 2000; Vörösmarty et al., 2010). Despite the small area they occupy, rivers are hotspots for human development, which puts immense pressure on the biodiversity that depends on them (Strayer & Dudgeon, 2010). Main threats include water pollution, overexploitation, modification of flow, alien species invasions, and habitat fragmentation or destruction (Vörösmarty et al., 2005; Dudgeon et al., 2006). Together, these impacts are so severe that >20% of freshwater species are either extinct or under threat of extinction (Abramovitz, 1996; Strayer & Dudgeon, 2010).

Various criteria exist to prioritize sites for conservation action. Some examples include sites with many threatened species, or with a large number of narrow range endemics, or regions that are extremely biodiverse, or that have high genetic uniqueness. The Greater Cape Floristic Region (GCFR) of South Africa is a biodiversity hotspot, and one of six floral kingdoms (Pressey et al., 2003; Born et al., 2007). The region is a proclaimed Global 200 Freshwater Ecoregion, as the degree of diversity and endemism for aquatic invertebrates is comparable to that of its terrestrial plants (Wishart & Day, 2002). Therefore, conserving GCFR rivers should be a priority so as to protect the large number of endemic species. However, numerous limitations, such as time, personnel, resources, and insufficient knowledge of species requirements, hamper conservation efforts in biodiversity hotspots like the GCFR. Conservation biologists often depend on the development of quick and effective ways to assess biodiversity and ecosystem health (Bani et al., 2002; Roberge & Angelstam, 2004). The use of surrogates for whole-ecosystem assessment and monitoring is a popular but controversial research field in conservation biology (Lindenmayer et al., 2015; Hunter et al., 2016). Identification of a species or group of species that can successfully represent various other components of an ecosystem allows for rapid conservation action and saves on human resources.

Aquatic invertebrates are the most widely used surrogate organisms in freshwater assessments (Bonada et al., 2006). Of the taxa used, the Odonata (dragonflies and damselflies) comprise some of the most common and sensitive species associated with rivers (Catling, 2005). The

group also contains numerous regional endemics, with the GCFR considered a centre of endemism for South African Odonata (Samways & Sharratt, 2010; Samways et al., 2011). This ancient order of well-studied insects can be strongly affected by water quality (Clausnitzer, 2003; Catling, 2005; Stewart & Samways, 2008; Kietzka et al., 2017). Adult male individuals are ideal candidate indicators, as they are highly territorial, with colourful, conspicuous bodies, and are easy to identify to species level (Simaika & Samways, 2009a; Futahashi et al., 2015; Samways & Simaika, 2016). They are widely used as indicator surrogates for water quality (Simaika et al., 2016; Valente-Neto et al., 2016; de Moor, 2017; Lesch & Bouwman, 2018), and although they have been suggested as umbrella species (Kim et al., 2016), their potential has only been shown for wetland plants in the USA (Bried et al., 2007). The success of Odonata species as indicators even led to the development of the Dragonfly Biotic Index (DBI), which has been used for water quality assessments, conservation monitoring, and in reserve selection in South Africa (Simaika & Samways, 2009a, b).

In addition to the Odonata, the larvae of the taxon set Ephemeroptera (mayflies), Plecoptera (stoneflies) and Trichoptera (caddisflies) (collectively known as the EPT) have been widely used as indicators of water quality (Paparisto et al., 2009). The heterogeneous nature of the GCFR mountainous headwaters provide a range of unique habitat conditions. These are required to sustain many of the rare and endemic species from these groups. For example, a few specialist EPT species are limited to single rivers (Dallas & Day, 2007), and are also sensitive to changes in water quality, making them important components in water quality assessments, such as the EPT Biotic Index and the South African Scoring System (SASS) (Lenat, 1988; Lenat & Crawford, 1994; Haggag et al., 2018). Furthermore, comparisons between aquatic macroinvertebrate scores and DBI scores are correlated (Magoba & Samways, 2010).

Assessments aim to have a range of scores, and when high, show that water quality is good, but when low, that water quality is low. However, the presence of environmentally sensitive taxa (and good water quality) does not necessarily mean that there are high levels of biodiversity at that particular site (i.e. these are not measures of high biodiversity). By prioritizing these specific sites for conservation action, there is no guarantee that the conserved sites will maintain high biodiversity. Therefore, it is crucial to also identify ways in which to successfully measure and conserve biodiversity using appropriate surrogates.

The umbrella species concept is a biodiversity surrogacy method that aims to conserve a large number of species in an ecosystem, by focusing on a single species or a select group of co-occurring species (Fleishman et al., 2000; Bried et al., 2007). This approach is management-orientated, and uses surrogate species to assist in making conservation-related decisions. The concept contrasts with that of biodiversity indicators where surrogate species richness defines the species richness of other taxa in the same area (be it high or low) (McGeoch, 1998; Fleishman et al., 2000). Successful surrogate species can also assist in the allocation of scarce resources, and to prioritize sites that require conservation action (Fleishman et al., 2000; Bried et al., 2007). Initially, large vertebrates with large area requirements were suggested as the best potential surrogates, on the premise that if enough space was provided for these species, many other species with smaller area requirements would be conserved (McNab, 1963; Lambeck, 1997; Caro, 2003). However, this approach overlooks differences in species abundance and richness, does not acknowledge individual species requirements (especially at smaller scales), can easily exclude local diversity hotspots (Kerr, 1997), and risks of range retraction and even extinction of the surrogate. In addition, the setting aside of such massive areas of protected land is, in many cases, unrealistic, and not always beneficial to river ecosystems, when the main focus is on terrestrial biota.

The concept of an umbrella (i.e. surrogate) includes any species or group of species whose conservation results in the protection of co-occurring species (Lambeck, 1997). The notion behind the concept is credible, but its feasibility criticized, without a method to quantify the selection of surrogate species (Branton & Richardson, 2011). Therefore, the umbrella index was developed as a quantitative method to select the best surrogate species or taxon set for representing certain other species or taxon sets (Fleishman et al., 2000; 2001). For each potential surrogate species evaluated, a score is calculated based on a combination of its co-occurrence with other species (number of sites where both occur), its occurrence rate (number of sites that this species occupies), and its sensitivity to anthropogenic disturbance (Fleishman et al., 2000; 2001). Therefore, based on this emended definition, a successful surrogate species should often occur with the other taxa that are to be conserved, it must have a moderate occurrence rate within sites (ideally 50% site occupancy to optimize sampling intensity), and it must be more sensitive to human disturbance than most of the other taxa (Fleishman et al., 2000; Bried et al., 2007). To date, the umbrella index has only been applied to birds and butterflies (Betrus et al., 2005), and to Odonata and wetland plants in the USA (Bried et al., 2007).

We aim here to evaluate the performance of the surrogate species approach using the umbrella index as a tool for biodiversity conservation in an area with many rare and threatened freshwater species. We also determine whether Odonata and EPT species respond similarly to environmental variables, as both groups are known to be sensitive to changes in their habitats. We expect to identify surrogate species in each group that are significantly better management surrogates for their own group (Odonata surrogates for overall Odonata, and EPT surrogates for overall EPT) than for the other group (Odonata surrogates for EPT, and EPT surrogates for Odonata). If one group acts as a good surrogate for taxa in the other group, then this will allow measurement of a greater proportion of the overall assemblage with more efficiency. Furthermore, if either group could be successfully used as both environmental indicators and biodiversity surrogates in conservation planning, this would allow for the prioritization of sites for high water quality and biodiversity conservation.

3.2 Materials and methods

3.2.1 Study area

Study sites were selected along four rivers in the Greater Cape Floristic Region (GCFR), Western Cape Province, South Africa. The area has a Mediterranean climate, with natural vegetation consisting mostly of sclerophyllous fynbos on quarzitic sandstone. Rivers here are characteristically acidic and low in dissolved solids and nutrients, and are physically complex (Day, 1995; Samways et al., 2011). The upper reaches of these small rivers are usually subjected to the least anthropogenic influences, and are often inhabited by sensitive, endemic species (Palmer et al., 2005). These rocky sections are turbulent and low in discharge, but as rivers progress to middle/ lower reaches, they broaden and become less turbulent, with pebbled or sandy beds. Here, they are often affected by anthropogenic disturbance related to agriculture or urbanization (de Moor & Day, 2013).

The selected sites comprised sections of the Eerste (34°00'28.19"S; 19°00'6.46"E), Lourens (34°00'56.90"S; 18°58'42.54"E), Palmiet (34°03'24.95"S; 19°00'48.52"E) and Molenaars (33°42'00"S; 19°13'60"E) Rivers, which were chosen for having large areas of natural, untransformed land, as well as agriculturally transformed land (into vineyards and orchards) (Fig. 3.1). For the Eerste, Lourens and Molenaars Rivers, natural sites were confined to the upper river reaches, and the agricultural sites to the middle/ lower reaches. The Palmiet River,

differs from the other three rivers, as its untransformed section occurs in the lower reaches, with the upper reaches transformed by agriculture.

For each river, 45 natural and 45 agricultural sites were randomly selected using QGIS 2.14.3 (QGIS Development Team, 2014). Each site consisted of a 10 x 3 m transect of river, which ran parallel to the river's bank. Sampling took place twice per site, once during spring/early summer and again in autumn, to account for species seasonal differences. This is according to recommendations from Grant and Samways (2007) for Odonata species of the Western Cape Province. Similarly, season also has an influence on macroinvertebrates in the Western Cape. According to Dallas (2004), higher species richness of macroinvertebrates occurs during spring but a greater number of sensitive species are present during autumn. Sites were equally divided between three habitat types, defined by flow turbulence, and included deposition pools (P) of still water (< 0.01 m/sec), glides (G) with low turbulence (surface flow 0.01 - 0.4 m/sec), and riffles (R) with high turbulence (surface flow > 0.4 m/sec).

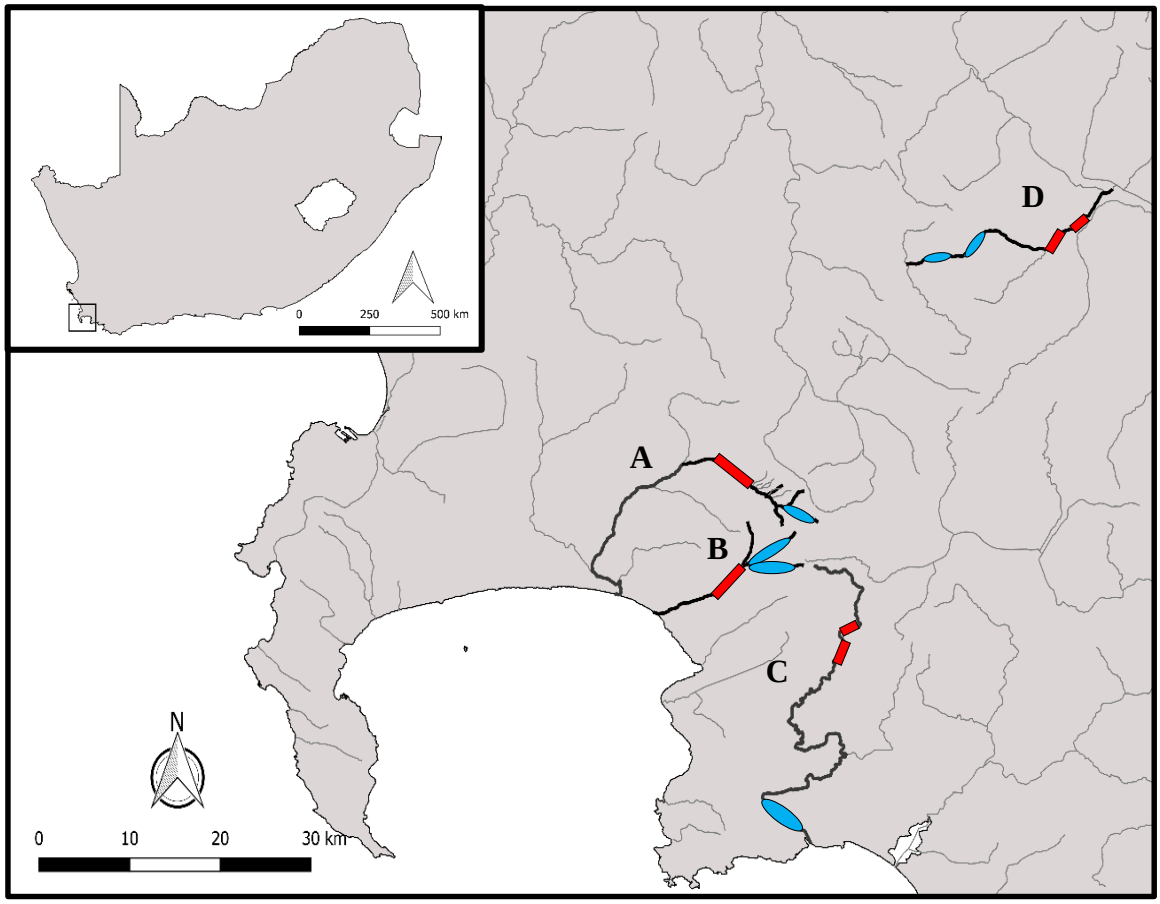


Fig. 3.1 Map showing the country of South Africa (top left) and the four focal Western Cape Province rivers. A) Eerste River, B) Lourens River, C) Palmiet River, D) Molenaars River. Blue ovals indicate sections of the rivers where the natural sites occurred (45 sites per river) and red rectangles indicate areas where the agricultural sites occurred (45 sites per river).

3.2.2 Environmental variables

At each site, over two seasons, 24 environmental variables (EVs) were recorded. These included the categorical variables of river catchment (Eerste, Lourens, Palmiet, or Molenaars), land use (natural or agricultural) and habitat type (pool, glide, or riffle). The degree of shading was estimated as the percentage of shade over the river (excluding river banks) at midday for each site. Due to shade cover varying throughout the day, observations were only conducted at midday. The average width and depth of the river at each site was measured with a tape measure.

Elevation and site positions were recorded with a handheld GPS. Water parameters (temperature, dissolved oxygen, conductivity, and pH) were measured using a multi-parameter water quality meter (Model: YSI 556 Multi Probe System; Make: YSI Fondriest Environmental). For the rivers' edges (10 x 1.5 m of land), the percentage of boulders, rocks, sand, and detritus were visually estimated by two observers. The same variables were estimated for the rivers' beds for the other, in-water part of the measured site (10 x 1.5 m of river). For vegetation data, the percentage cover and average height of both alien and indigenous vegetation were estimated (trees, shrubs, grass, and aquatic macrophytes), and used to determine average indigenous and exotic cover per entire site (10 x 3 m). The percentage of *Prionium serratum* (Palmiet Reed) was recorded as a separate variable, as it often dominates natural areas and positively influences Odonata diversity (Samways & Sharratt, 2010).

3.2.3 Odonata sampling

Adult male dragonflies were counted by two observers for 30 min per site between 10:00 and 15:00 on windless, sunny (< 10% cloud cover) days, when dragonfly activity is at its highest. Visual scanning was used, as it has previously been found to be 100% accurate for Anisoptera and 80% accurate for Zygoptera, which was made even more accurate by using close-focus binoculars when needed (Moore, 1991; Suh & Samways, 2005). Male Odonata are territorial, and therefore largely site-faithful, which makes observing them easy (Switzer, 1997). To avoid duplication of counts, or when identification was difficult, specimens were collected, identified

in-hand using Samways and Simaika (2016), and kept as voucher specimens or released after being identified.

3.2.4 Ephemeroptera, Plecoptera and Trichoptera sampling

EPT larvae are aquatic, and were sampled using similar protocol to that used for SASS5 (Dickens & Graham, 2002). All available microhabitats were sampled at each site to account for biotope preferences of species, which included stones (in-current and out-of-current), vegetation (marginal and aquatic vegetation), and sediment (gravel, sand and mud). The kick-method was used, where stones and sediment are disturbed by foot, and a 30 cm x 30 cm, 1000 µm mesh net submerged to collect samples for 3 min per site. Collected individuals were emptied into a sorting tray, and all EPT individuals separated, and preserved in 90% ethanol on site. Samples were later sorted to morphospecies, and identified to family level.

3.2.5 Data analyses

3.2.5.1 Species richness and abundance

EstimateS version 9.1 (Colwell, 2013) was used to determine whether sampling effort was sufficient. The non-parametric species estimators ICE and Chao2 were calculated, and samples were randomised 999 times. Non-parametric estimators are generally used for invertebrate assemblages, especially in regions like the GCFR, where a large number of endemic species in low abundance occur (Hortal et al., 2006).

3.2.5.2 Identification of potential surrogate species

The umbrella index was used to determine the potential of each species to act as a surrogate for the overall species of the same group (EPT for EPT; Odonata for Odonata). Calculations were done separately for EPT and Odonata groups. The index is based on the sum of three sub-indices: 1) the mean percentage of co-occurring species, 2) degree of ubiquity, and 3) sensitivity to human disturbance (Fleishman et al., 2001). These indices were calculated as follows:

1) Mean percentage of co-occurring species (PCS) is measured on a scale from 0 (occurs with few other species) to 1 (occurs with many other species). For each species j, PCS is calculated as:

$$PCS = \sum_{i=1}^{l}[S_i - 1)/(S_{max} - 1)]/Nj$$

Where l is equal to the number of sites in the data set, S_i is the number of species present at each site i, S_{max} is the highest possible number of species that can be present at a site, and Nj is the number of sites at which species j is present.

2) The median rarity (*R*) for each species *j* is calculated as:
$$R = 1 - |0.5 - Qj|$$

Where *Qj* = 1- (*N*present/*N*total) and *N* is number of sites (Baz, 1991). R ranges from 0.5 to 1 with species that are very rare or common are closer to 0.5, while species that have an intermediate degree of ubiquity are equal to 1.

3) Sensitivity to human disturbance (DSI) are taxon- and ecoregion-specific, and quantified on a relative scale ranging from 0 (low sensitivity) to 1 (high sensitivity). For dragonfly species, the DSI score was extracted from the Dragonfly Biotic Index (DBI), where sensitivity to human disturbance is one of the sub-indices (Samways & Simaika, 2016). For the EPT taxa, sensitivity scores were scaled from the South African Scoring System (SASS), a water quality index (Dickens & Graham, 2002).

   DSI = (DBI or SASS score) / max allowable (DBI/SASS score)

The umbrella index for each species was calculated as:

$$UI = PCS + R + DSI$$

From these scores, the groups of potential surrogate species were identified as those with a UI exceeding the mean UI for the group plus one standard deviation.

3.2.5.3 Cross-group surrogate potential

McNemar's *Q*-test (1974) was used to compare the proportion of species protected when using surrogate species from the same group (EPT surrogates for overall EPT; Odonata surrogates for overall Odonata) vs. for the opposite group (EPT surrogates for Odonata overall; Odonata surrogates for EPT overall). A non-parametric Chi-square test was used to determine differences between count data and dependent proportions (Agresti, 1992). Discordant pairs

had greater power when they have similar species proportions, compared to comparisons with dissimilar species proportions (Wacholder & Weinberg, 1982). Odonata and EPT proportions were sorted into a 2×2 contingency table of the selected surrogate species vs. the beneficiary group (either the overall assemblage from the same group, or the overall assemblage from the opposite group). Comparisons were conducted each at 20% (72 sites), 50% (180 sites) and 80% (288 sites), which included the sites where the highest number of surrogate species were present, this is reffered to as effort in the results. Using various sampling efforts enabled us to find the balance between time-and-effort and interpretability of data, and for future study recommendations. Analyses were carried out within R software (R Core Team, 2018), using the *rcompanion* package.

Richness covariance between EPT taxa and Odonata was tested using linear regressions conducted in R software (R Core Team, 2018) using the *lme4* package (Bates & Sarkar, 2008). Mantel tests were also conducted in R software (R Core Team, 2018), using the *ade* package (Chessel et al., 2011). This tested the compositional similarity levels between EPT taxa and Odonata, using dissimilarity matrices of Sørensen coefficient values. Pair combinations of sites were tested against 999 randomized correlations, which determined whether non-random associations existed between taxa. High-variability in our data could result in a significant trend ($P < 0.05$), but have low correlation coefficient. This would imply that the data is noisy but the trend is still significant even though data points are scattered far from the regression line. Some studies found low correlation in species richness among taxa at small scales associated with local management (Prendergast et al., 1993; Chase et al., 1998; Rubinoff, 2001; Betrus et al., 2005). Despite this, assessments between taxa at larger scales are more unequivocal (Olson & Dinerstein, 1998; Ricketts et al., 1999; Stein et al., 2000; Betrus et al., 2005).

3.2.5.4 Factors influencing species richness

For species richness of overall EPT, EPT surrogates, overall Odonata, and Odonata surrogates, best fit models were determined with a stepwise forward selection method based on Akaike Information Criteria (AIC) values using the *AICcmodavg* package (Mazerolle & Mazerolle, 2017). As species richness data were non-normal, thereafter, generalized linear mixed-effect models (GLMMs) were used to identify which environmental variables were the most important for determining species richness. These analyses were performed using the *lme4* package (Bates et al., 2014) in R (R Core Team, 2018). In all instances, GLMMs with a Laplace

approximation and a Poisson distribution were used (Bolker et al., 2009). When tested, no over dispersion of variances was shown, and thus, χ^2- and P-values were calculated (Bolker et al., 2009). River identity was included as the random effect, and the remaining variables were included as fixed effects. For significant, categorical variables, Tukey *post hoc* tests in the *R* package *multcomp* were used to determine pairwise differences (Hothorn et al., 2008).

3.3 Results

3.3.1 Species abundance and richness

A total of 11 130 Ephemeroptera (6 families, 17 morphospecies), 611 Plecoptera (1 family, 3 morphospecies), 4 038 Trichoptera (9 families, 39 morphospecies) and 1 268 Odonata (8 families, 26 species) individuals were sampled. Overall, each order rarefaction curve reached an asymptote or near-asymptote (Appendix 3.A). This indicates that adequate sampling was conducted and was sufficient and reflective of the species present. Additionally, this was supported by species estimates for Ephemeroptera (Chao2 = 16 (0.35) and ICE = 16), Plecoptera (Chao2 = 3 (0) and ICE = 3), Trichoptera (Chao2 = 44.99 (5.53) and ICE = 43.78) and Odonata (Chao2 = 27.99 (4.17) and ICE = 28.93).

3.3.2 Umbrella index scores

For EPT taxa, the umbrella index scores of eight EPT species ranked above the minimum score (1.46) for surrogate potential (Fig. 3.2; Table 3.1). The top-ranked EPT surrogate species was from the order Plecoptera and the family Notonemouridae, and overlapped with 36% of total EPT occurrence. When all the EPT surrogate species were included, they overlapped with 88% of total EPT occurrence. For the Odonata, seven surrogate species fell above the cut-off point (1.55) (Fig. 3.2; Table 3.1). The top-ranked Odonata surrogate, *Pseudagrion furcigerum*, overlapped with 17% of total Odonata species occurrence. When all seven surrogate species were included, they overlapped with 77% of total Odonata occurrence. Although high, neither the EPT, nor the Odonata surrogate schemes, reached 100% site representation (saturation).

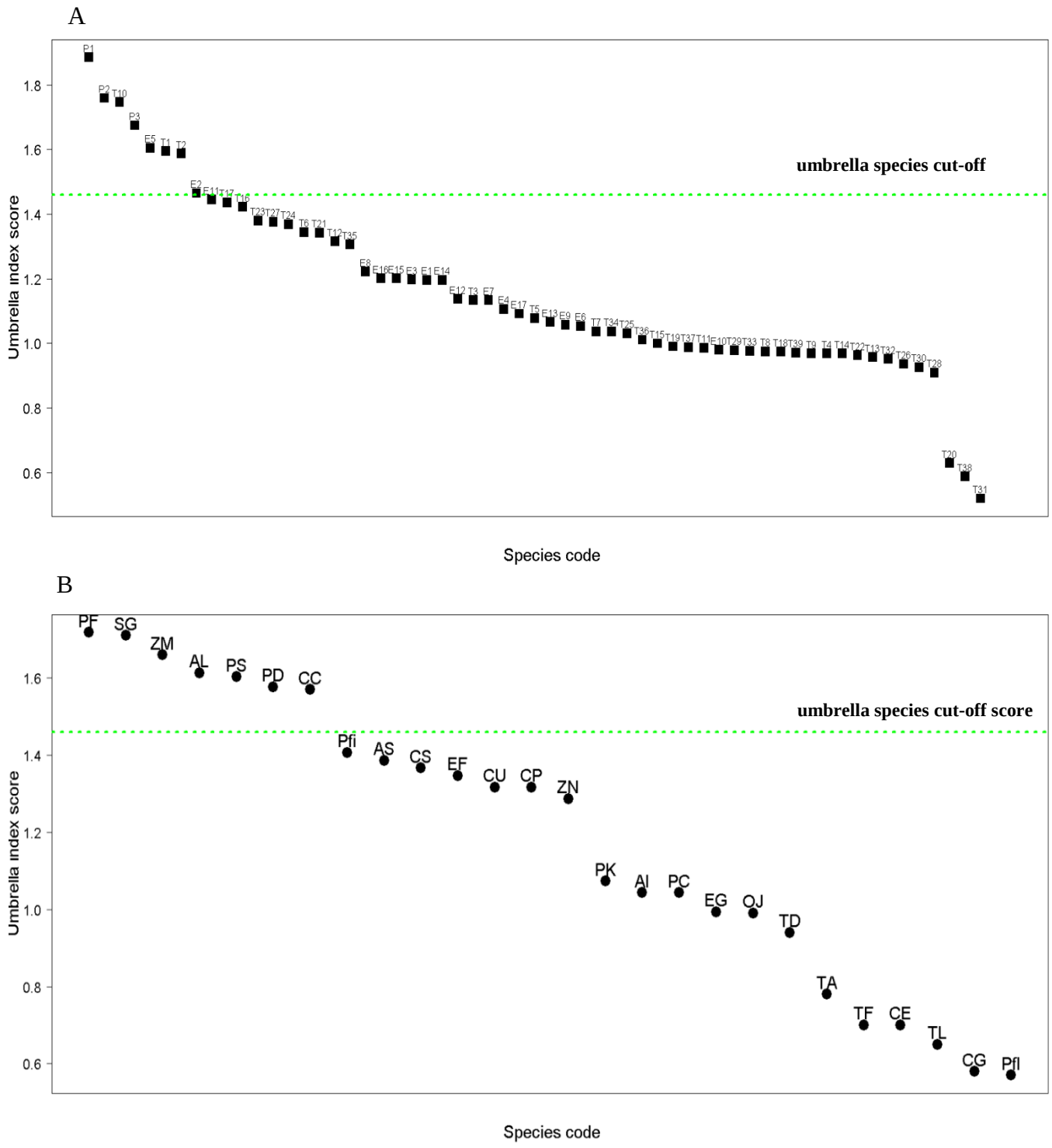


Fig. 3.2 Scatterplots of the umbrella index scores (UIS) of A) Ephemeroptera/ Plecoptera/ Trichoptera (EPT) taxa, and B) Odonata. Dotted line represents the minimum score required to be considered a surrogate species (Mean UIS + 1 SD). For A) letters: E = Ephemeroptera, P = Plecoptera and T = Trichoptera. Numbers alongside E/P/T letters denote the allocated morphospecies numbers. For B) please see Appendix 3.B for descriptions of species codes for Odonata.

Table 3.1 Species codes, ranked umbrella index (UI) scores, additive proportion of river sites containing top surrogate species (effort) and additive response currency for Ephemeroptera/ Plecoptera/ Trichoptera (EPT) and Odonata. TO = the total occurrences (the total incidence of all species in a given surrogate scheme). Letters: E = Ephemeroptera, P = Plecoptera and T = Trichoptera. Numbers alongside E/P/T letters denote the allocated morphospecies numbers.

	Code	UI score	Cumulative TO %
EPT			
Notonemouridae sp.2	P1	1.885	36
Notonemouridae sp.3	P2	1.759	39
Hydrosalpingidae sp.1	T10	1.747	48
Notonemouridae sp.4	P3	1.675	51
Teloganodidae sp.1	E5	1.605	71
Philopotamidae sp.1	T1	1.595	81
Barbarochthonidae sp.1	T2	1.587	85
Heptageniidae sp.1	E2	1.465	88
Odonata			
Pseudagrion furcigerum	PF	1.718	17
Syncordulia gracilis	SG	1.710	19
Zosteraeschna minuscula	ZM	1.660	21
Allocnemis leucosticta	AL	1.613	25
Pinheyschna subpupillata	PS	1.609	60
Pseudagrion draconis	PD	1.577	76
Chlorolestes conspicuus	CC	1.570	77

3.3.3 Same-group vs. cross-group surrogate potential

No significant differences were detected between protection levels of same-group vs. cross-group surrogate schemes for EPT or Odonata (Table 3.2). At 80% effort, EPT surrogate species protected 98% of overall EPT species, and 100% of the Odonata species present. At 80% effort, Odonata surrogate species protected 100% of the Odonata and EPT species present. Cross-group patterns of species richness were linear ($r^2 = 0.07$, $P < 0{\cdot}001$), and non-random patterns of association were found by the multivariate correlation ($r = 0.043$, $P < 0.01$). Sites shared between surrogate species at the different sampling efforts are given in Appendix 3.C.

Table 3.2 Proportion of all possible river site combinations with response currency (total occurrences) equal to, or exceeding that, of given surrogate schemes. The surrogate schemes for both groups included the top percentage (effort) of sites containing the most potential surrogate species. U same = surrogates and taxa from the same group; U cross = surrogates and taxa from opposite groups. EPT = Ephemeroptera/Plecoptera/Trichoptera.

Surrogate scheme	**Proportion of species protected**	
	U same	**U cross**
EPT		
72 sites (20% effort)	0.80	0.69
180 sites (50% effort)	0.87	0.92
288 sites (80% effort)	0.98	1.00
Odonata		
72 sites (20% effort)	0.92	0.75
180 sites (50% effort)	0.96	0.93
288 sites (80% effort)	1.00	1.00

3.3.4 Factors influencing species richness

For all the groups, species richness significantly differed between the land use types (Table 3.3). For EPT overall and surrogates, and for Odonata overall, significantly more species occurred at natural sites, whereas Odonata surrogate species richness was higher at agricultural sites (Appendix 3.D). At deposition pools in the rivers, significantly fewer EPT surrogate species were recorded, whereas overall Odonata species richness was at its highest (Appendix 3.E). Land use and water pH were the most influential variables for EPT overall and surrogate species richness (Table 3.3). Water temperature affected all groups, and was the most influential variable for Odonata overall and Odonata surrogate species (Table 3.3). There were fewer EPT species but a greater number of Odonata species, at sites with higher water temperatures (Appendix 3.F). There were also fewer overall and surrogate EPT species at sites with higher pH levels, but higher overall Odonata species richness (Appendix 3.F).

Table 3.3 Effects of environmental variables on species richness for Ephemeroptera/Plecoptera/Trichoptera (EPT) and Odonata. Test-statistics are displayed as χ^2 values and significant values are in bold text. Environmental variables not included in the best fit models are marked with a -.

Variables	χ^2 values			
	EPT	**EPT surrogates**	**Odonata**	**Odonata surrogates**
Land use	**30.50*** **	**33.31*** **	**10.52** **	**6.00***
Flow type	3.87	**19.93*** **	**10.26** **	-
% shade	1.41	-	-	-
Width	-	-	-	-
Depth	-	-	**12.30*** **	-
Water measures				
Water temperature	**8.98** **	**5.16***	**31.97*** **	**23.08*** **
Dissolved oxygen	-	-	-	-
Conductivity	-	-	-	-
pH	**40.46*** **	**32.63*** **	**7.34** **	-
River edge cover				
% debris	-	-	-	-
% sand	-	-	-	-
% rock	-	-	-	-
% boulder	-	-	-	-
River bed cover	-			
% debris	-	**16.70*** **	**4.27***	-
% sand	**7.07** **	-	-	-
% rock	-	-	-	-
% boulder	-	-	-	-
Vegetation				
Avge veg height	-	-	-	-
% indigenous cover	-	-	-	-
% alien cover	-	-	-	-
Prionium serratum	-	-	-	-
Aquatic macrophytes	-	-	-	2.66

* P < 0.05, ** P < 0.01, *** P < 0.001

3.4 Discussion

We show that a selection of a few surrogate species from both groups are not just good surrogates for the protection of their own taxon or taxon group, but that they are also good surrogate taxa for each other. At 80% effort, the Odonata surrogates were able to cover all of the Odonata species, and all of the EPT species encountered. Species richness of all four groups were sensitive to changes in habitats, and were influenced largely by the same environmental variables. Therefore, by protecting the surrogate species of any one group identified here, it would be possible not only to protect that specific taxon/taxon group, but also possibly a large proportion of the local biodiversity, as the surrogate species are highly sensitive taxa. This would also cover the natural variability between four different rivers, including all the different biotopes encountered.

We found that Odonata and EPT are both good surrogate taxa for conservation planning in the GCFR. However, when surrogate species from the same group were used, the proportion of EPT protected was slightly lower than the proportion of Odonata. The proportion of protected species was not reduced for either group by using cross-group surrogate species. In fact, Odonata surrogate species protected a slightly higher proportion of EPT overall (Odonata surrogates protected 100% of overall EPT at 80% effort) than the EPT surrogates did for their own group (EPT surrogates protected 98% of overall EPT at 80% effort). In other words, EPT surrogates protected a higher proportion of the Odonata overall (EPT surrogate protected 100% of overall Odonata at 80% effort) than of their own assemblage (EPT surrogate protected 98% of overall EPT at 80% effort). This may reflect that species turnover of EPT tends to be greater than that of Odonata, which may be due to their great diversity, limited dispersal abilities or them having very specific microhabitat requirements. Many adult Odonata have greater dispersal abilities than EPT taxa, with some Anisoptera species being strong fliers, and can travel far distances to select favourable habitat conditions (May, 1976, 1982; McGeoch & Samways, 1991). In the case of *Pantala flavescens*, which is exceptional, it has been recorded to fly more than 6 000 km, making it the farthest flying insect (Hobson et al., 2012). Even as winged adults, EPT taxa are weaker fliers. Furthermore, some species, as in the Ephemeroptera, are extremely short-lived as adults, which limits their time for dispersing far, making it difficult for them to overcome landscape barriers (Pearson & Boyero, 2009). Some researchers have reported that adult Ephemeroptera and Trichoptera are typically recorded within 50 m of a river or stream (Jackson & Resh 1989; Collier & Smith 1997; Petersen et al., 1999; 2004). Although

incomparable to distances covered by some adult Odonata, many New Zealand Trichoptera can disperse up to 1 500 m (Graham et al., 2017). For the EPT larvae, average traveling distances were found to be between 0 and 200 m (Graham et al., 2017).

The species richness of these EPTO groups also responded to very similar environmental variables, with the most important drivers measured here being land use, flow type, and in-water measures. These are important variables for water quality, highlighting the ability of these groups to act as indicators of anthropogenic impacts and water quality (Dickens & Graham, 2002; Kietzka et al., 2017). This means that not only do EPT and Odonata co-occur, but that their richness is also driven by similar environmental variables. Thus, conservation efforts focussed on one of the groups would ultimately conserve the important areas and variables for the other.

As the umbrella index selects species with a median occurrence and that are habitat sensitive, this means that despite rivers in the GCFR being naturally heterogeneous (Day, 1995), with each having its own unique macroinvertebrate assemblage (Schael & King, 2005), we were able to successfully identify one suite of surrogate species for both EPT and Odonata assemblages across this region for both untransformed and agricultural sections. The suite of potential surrogate species were identified as those with an umbrella index score exceeding the mean score for the group plus one standard deviation. Some species were only present in one of the four rivers, and the umbrella index allowed for the protection of these rarer species by including them as surrogates. Although these species would have scored lower in terms of the medium rarity sub-index, they had high scores for the sensitivity to human disturbance sub-index. This was the case for the four highest scoring EPT surrogates that were from the Notonemouridae (three morphospecies) and Hydrosalpingidae (one morphospecies) families, and for the Odonata surrogate species *Syncordulia gracilis, Zosteraeschna minuscula* and *Chlorolestes conspicuus* which fell into this same group.

Some surrogates, such as the Odonata species *Pinheyschna subpupillata* and *Pseudagrion draconis*, were less sensitive to human disturbance, but had high scores as they co-occurred with many other species, and were neither common nor rare (i.e. of median rarity). All of the selected EPT surrogates had similar scores for the mean percentage of species with which they co-occurred. The last four EPT surrogate species were from the families Teloganodidae, Philopotamidae, Barbarochthonidae and Heptageniidae, and although still sensitive to human

disturbance, were less sensitive than the above-mentioned EPT surrogates but were closer to median rarity.

Lastly, some Odonata surrogates were selected for being sensitive to human disturbance, occurring with a many other species, and being somewhere between rare and median rarity. In this group were *P. furcigerum* and *Allocnemis leucosticta*. This way of scoring allowed for the selection of a suite of surrogate species ranging from some rare, highly sensitive endemics, which co-occurred with many other species to some that were more common, less sensitive species that co-occurred with many other species.

Overall we found that the umbrella index functioned well, even in a region with high endemism levels. This is important for conservation management, because where urgent action is required, the approach could aid in the rapid selection of sites across the region. If conservationists were to conserve the top 50% of sites where the most surrogate species were present, the EPT surrogates would conserve 87% of all the EPT species, and 92% of all the Odonata species. For the Odonata surrogates 96% of all the Odonata species would be conserved, and 93% of all the EPT species. Other umbrella studies also reported adequate to high protection levels (Fleishman et al., 2000; 2001). Betrus et al. (2005) used the umbrella index, and found for butterflies and birds in the USA, equal proportions of species would be conserved when selected umbrella species from the same group or opposite taxon group were used. Additionally, results from Bried et al. (2007) in USA wetlands, suggested transferability of Odonata and plant umbrella schemes. However, relative to random and non-umbrella species schemes, those using selected umbrella species did not significantly improve, but rather provided similar conservation returns. Despite this, they endorsed the use of the umbrella index for future quantitative selection of umbrella species, highlighting that some assemblages, depending on the area, ecosystem and circumstances involved, may not comprise species that satisfy index criteria.

We show here that the umbrella index developed by Fleishman et al. (2000) is an effective method for identifying prospective surrogate taxa for rivers with varying degrees of anthropogenic disturbance in a biodiversity hotspot. The development of such a method is in response to limited resources and urgent conservation challenges (Fleishman et al., 2001; Betrus et al., 2005). The umbrella index has proven its use not only in our complex,

biodiversity-rich region, but also for rivers, which are renowned for their heterogeneous and sensitive nature.

The great flexibility of the umbrella index for different taxa and ecoregions is evident here, which was one of the main motives behind its development (Fleishman et al., 2000). Furthermore, its ability to perform across a spatial scale greater than a single river is a particularly important criterion when the index is to be used in future conservation planning. This flexibility was also presented by Betrus et al. (2005), as umbrella species selected and applied to their own mountain range showed to be equally as effective when applied to a different mountain range. A similar conclusion was reached by Bried et al. (2007) for Odonata and vascular plants between different wetland sites. Despite its success shown here, the construction of a method for surrogate species selection that can be used in actual conservation planning would require the umbrella index to be tested widely for various taxa, ecoregions, and at different spatial scales.

The spatial associations between EPT and Odonata groups suggest that conservation managers could choose either one for adequate conservation planning (Clausnitzer, 2003; Schael & King, 2005; Samways, 2008; Robson et al., 2013). The choice of which group to use would depend on the specific question asked, and other considerations such as the amount of resources and personnel available, the time of year, time constraints, and level of taxonomic experience. Adult male Odonata are colourful, conspicuous and easy to locate at their riverside territories (Suhling et al., 2015; Moore & Martin, 2018). They are quick to sample, requiring only a net, and are easy to identify to species level with the use a field guide (Samways & Simaika, 2016). Therefore, in terms of approachability and effort, Odonata are the more easily measured assemblage, and certainly would be the more suitable choice for field workers in general, especially citizen scientists. However, although the Odonata surrogate taxon set was a slightly better representative of aquatic insect diversity recorded here, the use of particular Odonate taxon sets would need to be tested elsewhere to redefine the taxon set to match species turnover across the region, especially as the narrow-range endemics will change greatly with increasing distance across the region. Furthermore, Odonata studies work best in summer, as this is their peak activity season, and only on sunny, windless days at certain daylight hours. Any overwintering adults are only a small sub-set of the total number of species in any one area (van Schalkwyk et al., 2014), making it essential to understand the comparative phenology of the various species when making a full assessment (Samways & Grant, 2007). In contrast,

assessors who require urgent or rapid biodiversity surveys at any time of the year, and during almost any weather conditions, may prefer to use EPT larvae. This group here reached an asymptote sooner than Odonata, and thus surveys could be conducted at fewer sites. However, sampling of EPT has much greater challenges than sampling Odonata adults, and requires entering into the water and detailed post-sampling sorting which is far more tedious and difficult than identification of adult Odonata. Also, it is a greater taxonomic challenge to identify the larvae to species level. For this reason, EPT samples are usually identified only to family level in aquatic assessments, as in the South African Scoring System (SASS) (Dickens & Graham, 2002), and thus miss a lot of detail.

Here, we used two different sorts of taxon sets. For the EPT, we used morphospecies in family divisions, and for the Odonata, we identified individuals to species level. Of the three sub-indices in the umbrella index, only the sensitivity sub-index could potentially change if species-level scores were used instead of family-level scores. Unfortunately, this level of knowledge is not available for many taxa, including the EPT larvae here. For adult Odonata in South Africa, the value of conducting assessments at species level is evident from the accuracy at which the DBI operates at the microhabitat level (Simaika & Samways, 2009a; b). The robustness of the umbrella index is shown by the other two sub-indices (median rarity, and mean percentage of co-occurring species), which would remain unchanged whether used as morphospecies (family level) or identified to species level.

When applying the umbrella index to an ecosystem, priority should be given to taxa where species level information on sensitivity to human disturbance is available. Conservationists should aim to better quantify and rank a greater number of species based on their sensitivity to anthropogenic disturbances. This will not only allow us to determine which species could be at risk of extinction when faced with land transformation, but will also allow optimal performance of the umbrella index in numerous ecosystems and countries. Furthermore, identification of a group of successful surrogate species (to species level) in one area may also be applicable to the same type of ecosystem in close proximity to that area. Despite this, as seen here, when species level information is not available, using morphospecies and family level sensitivity information still successfully identified surrogate species, and should be considered in the absence of species level information.

3.5 Conclusion

The umbrella index can be applied in a biodiversity hotspot rich in endemic species, and to freshwater aquatic insect taxa that are widely used in freshwater assessments. With the limitation that Odonata adults can only be sampled in the summer, but with the positive attribute that they can be easily sampled and identified to the species level, means that they are the preferred surrogate. This was also borne out by the fact that EPT surrogates would conserve 87% of all the EPT species, and 92% of all the Odonata species, while the Odonata surrogates perform better, as they would conserve 96% of all the Odonata species, and 93% of all the EPT species at 50% effort.

3.6 References

Abramovitz, J. N. (1996). Imperiled waters, impoverished future : the decline of freshwater ecosystems. Washington D.C., Worldwatch Institute.

Agresti, A. (1992). A survey of exact inference for contingency tables. Statistical Science, 7(1), 131–153.

Bani, L., Baietto, M., Bottoni, L., & Massa, R. (2002). The use of focal species in designing a habitat network for a lowland area of Lombardy, Italy. Conservation Biology, 16(3), 826-831.

Bates, D., & Sarkar, D. (2008). The lme4 Package. URL http://cran. r-project. org.

Bates, D., Maechler, M., & Bolker, B. (2012). lme4: linear mixed-effects models using S4 classes. R package version 0.999375-42.

Baz, A. (1991). Ranking species and sites for butterfly conservation using presence–absence data in central Spain. Nota Lepidopterologica Supplement, 2, 4-12.

Betrus, C. J., Fleishman, E., & Blair, R. B. (2005). Cross-taxonomic potential and spatial transferability of an umbrella species index. Journal of Environmental Management, 74(1), 79–87.

Bolker, B. M., Brooks, M. E., Clark, C. J., Geange, S. W., Poulsen, J. R., Stevens, M. H. H., & White, J. S. S. (2009). Generalized linear mixed models: a practical guide for ecology and evolution. Trends in Ecology and Evolution, 24(3), 127–135.

Bonada, N., Prat, N., Resh, V. H., & Statzner, B. (2006). Developments in aquatic insect biomonitoring: A comparative analysis of recent approaches. Annual Review of Entomology, 51(1), 495–523.

Born, J., Linder, H. P., & Desmet, P. (2007). The Greater Cape Floristic Region. Journal of Biogeography, 34(1), 147–162.

Branton, M., & Richardson, J. S. (2011). Assessing the value of the umbrella-species concept for conservation planning with meta-analysis. Conservation Biology, 25(1), 9–20.

Bried, J. T., Herman, B. D., & Ervin, G. N. (2007). Umbrella potential of plants and dragonflies for wetland conservation: A quantitative case study using the umbrella index. Journal of Applied Ecology, 44(4), 833–842.

Caro, T. M. (2003). Umbrella species: critique and lessons from East Africa. Animal Conservation, 6(2), 171–181.

Catling, P. M. (2005). A potential for the use of dragonfly (Odonata) diversity as a bioindicator of the efficiency of sewage lagoons. Canadian Field-Naturalist, 119(2), 233–236.

Chase, M. K., Rotenberry, J. T., & Misenhelter, M. D. (1998). Is the California Gnatcatcher an indicator of bird-species richness in coastal sage scrub?. Western Birds, 29, 468-474.

Chessel, D., Dufour, A. B., & Dray, S. (2011). ade4: analysis of ecological data: exploratory and Euclidean methods in environmental sciences, version 1.4-17.

Clausnitzer, V. (2003). Dragonfly communities in coastal habitats of Kenya: Indication of biotope quality and the need of conservation measures. Biodiversity and Conservation, 12(2), 333–356.

Clausnitzer, V., Dijkstra, K. D. B., Koch, R., Boudot, J. P., Darwall, W. R., Kipping, J., Samraoui, B., Samways, M. J., Simaika, J. P., & Suhling, F. (2012). Focus on African freshwaters: hotspots of dragonfly diversity and conservation concern. Frontiers in Ecology and the Environment, 10(3), 129–134.

Collier, K. J., & Smith, B. J. (1998). Dispersal of adult caddisflies (Trichoptera) into forests alongside three New Zealand streams. Hydrobiologia, 361, 53-65.

Dallas, H. F. (2004). Seasonal variability of macroinvertebrate assemblages in two regions of South Africa: implications for aquatic bioassessment. African Journal of Aquatic Science, 29(2), 173–184.

Dallas, H. F., & Day, J. A. (2007). Natural variation in macroinvertebrate assemblages and the development of a biological banding system for interpreting bioassessment data - A preliminary evaluation using data from upland sites in the south-western Cape, South Africa. Hydrobiologia, 575(1), 231–244.

Day, J. M. (1995). Geographical patterns, and their origins, in the dominance of major ions in South African rivers. South African Journal of Science, 91(6), 299–306

de Moor, F. C. (2017). Dragonflies as indicators of aquatic ecosystem health. South African Journal of Science, 113, 1–2.

de Moor, F. C., & Day, J. A. (2013). Aquatic biodiversity in the Mediterranean region of South Africa. Hydrobiologia, 719(1), 237–268.

Dickens, C. W. S., & Graham, P. M. (2002). The South African scoring system (SASS) version 5 rapid bioassessment method for rivers. African Journal of Aquatic Science, 27(1), 1–10.

Dudgeon, D., Arthington, A. H., Gessner, M. O., Kawabata, Z. I., Knowler, D. J., Lévêque, C., Naiman, R. J., Prieur-Richard, A. H., Soto, D., Stiassny, M. L., & Sullivan, C. A. (2006). Freshwater biodiversity: importance, threats, status and conservation challenges. Biological Reviews of the Cambridge Philosophical Society, 81(2), 163–182.

Fleishman, E., Blair, R. B., & Murphy, D. D. (2001). Empirical validation of a method for umbrella species selection. Ecological Applications, 11(5), 1489–1501.

Fleishman, E., Murphy, D., & Brussard, P. F. (2000). A new method for selection of umbrella species for conservation planning. Ecological Applications, 10(2), 569–579.

Futahashi, R., Kawahara-Miki, R., Kinoshita, M., Yoshitake, K., Yajima, S., Arikawa, K., & Fukatsu, T. (2015). Extraordinary diversity of visual opsin genes in dragonflies. Proceedings of the National Academy of Sciences of the United States of America, 112(11), 1247-56.

Graham, N. (1999). Guidelines for Drinking-Water Quality, 2nd edition, Addendum to Volume 1 – Recommendations, World Health Organisation, Geneva, Urban Water, 1(2), 183.

Graham, S. E., Storey, R., & Smith, B. (2017). Dispersal distances of aquatic insects: upstream crawling by benthic EPT larvae and flight of sdult Trichoptera along valley floors. New Zealand Journal of Marine and Freshwater Research, 51, 146–64.

Haggag, A. A., Mahmoud, M. A., Bream, A. S., & Amer, M. S. (2018). Family variation of aquatic insects and water properties to assess freshwater quality in El-Mansouriya stream, Egypt. African Entomology, 26(1), 162–173.

Harrison, I. J., Green, P. A., Farrell, T. A., Juffe-Bignoli, D., Sáenz, L., & Vörösmarty, C. J. (2016). Protected areas and freshwater provisioning: a global assessment of freshwater provision, threats and management strategies to support human water security. Aquatic Conservation: Marine and Freshwater Ecosystems, 26, 103–120.

Hobson, K. A., Anderson, R. C., Soto, D. X., & Wassenaar, L. I. (2012). Isotopic evidence that dragonflies (*Pantala flavescens*) migrating through the Maldives come from the northern Indian subcontinent. PloS One, 7(12), 52594.

Hortal, J., Borges, P. A. V, & Gaspar, C. (2006). Evaluating the performance of species richness estimators: Sensitivity to sample grain size. Journal of Animal Ecology, 75(1), 274–287.

Hothorn, T., Bretz, F., & Westfall, P. (2008). Simultaneous Inference in General Parametric Models. Biometrical Journal, 50(3), 346–363.

Hunter, M., Westgate, M., Barton, P., Calhoun, A., Pierson, J., Tulloch, A., Beger, M., Branquinho, C., Caro, T., Gross, J. & Heino, J. (2016). Two roles for ecological surrogacy: Indicator surrogates and management surrogates. Ecological Indicators, 63, 121–125.

Jackson, J. K., & Resh, V. H. (1989). Distribution and abundance of adult aquatic insects in the forest adjacent to a northern California stream. Environmental Entomology, 18, 278–283.

Kerr, J. T. (1997). Species richness, endemism, and the choice of areas for conservation. Conservation Biology, 11(5), 1094–1100.

Kietzka, G. J., Pryke, J. S., & Samways, M. J. (2017). Aerial adult dragonflies are highly sensitive to in-water conditions across an ancient landscape. Diversity and Distributions, 23(1), 14–26.

Kim, J. H., Chung, H. Y., Kim, S. H., & Kim, J. G. (2016). The influence of water characteristics on the aquatic insect and plant assemblage in small irrigation ponds in Civilian Control Zone, Korea. Journal of Wetlands Research, 18(4), 331–341.

Lambeck, R. J. (1997). Focal species: A multi-species umbrella for nature conservation. Conservation Biology, 11(4), 849–856.

Lenat, D. R. (1988). Water quality assessment of streams using a qualitative collection method for benthic macroinvertebrates. Journal of the North American Benthological Society, 7(3), 222–233.

Lenat, D. R., & Crawford, J. K. (1994). Effects of land use on water quality and aquatic biota of three North Carolina Piedmont streams. Hydrobiologia, 294(3), 185–199.

Lesch, V., & Bouwman, H. (2018). Adult dragonflies are indicators of environmental metallic elements. Chemosphere, 209, 654–665.

Lindenmayer, D., Pierson, J., Barton, P., Beger, M., Branquinho, C., Calhoun, A., Caro, T., Greig, H., Gross, J., Heino, J., & Hunter, M. (2015). A new framework for selecting environmental surrogates. Science of the Total Environment, 538, 1029–1038.

Magoba, R. N., & Samways, M. J. (2010). Recovery of benthic macroinvertebrate and adult dragonfly assemblages in response to large scale removal of riparian invasive alien trees. Journal of Insect Conservation, 14(6), 627–636.

May, M. L. (1976). Thermoregulation and Adaptation to Temperature in Dragonflies (Odonata: Anisoptera). Ecological Monographs, 46(1), 1–32.

May, M. L. (1982). Heat exchange and endothermy in Protodonata. Evolution, 36(5), 1051.

Mazerolle, M. J., & Mazerolle, M. M. J. (2017). Package 'AICcmodavg'. R package.

McGeoch, M. A. (1998). The selection, testing and application of terrestrial insects as bioindicators. Biological Reviews, 73, 181–201.

McGeoch, M. A., & Samways, M. J. (1991). Dragonflies and the thermal landscape: implications for their conservation (Anisoptera). Odonatologica, 20(3), 303–320.

McNab, B. K. (1963). Bioenergetics and the determination of home range size. The American Naturalist, 97(894), 133–140.

Moore, M. P., & Martin, R. A. (2018). Trade-offs between larval survival and adult ornament development depend on predator regime in a territorial dragonfly. Oecologia, 188(1), 97-106.

Moore, N. W. (1991). The development of dragonfly communities and the consequences of territorial behaviour: a 27 year study on small ponds at Woodwalton Fen, Cambridgeshire, United Kingdom. Odonatologica, 20(2), 203–231.

Olson, D. M., & Dinerstein, E. (1998). The Global 200: a representation approach to conserving the Earth's most biologically valuable ecoregions. Conservation Biology, 12(3), 502-515.

Palmer, M. A., Bernhardt, E. S., Allan, J. D., Lake, P. S., Alexander, G., Brooks, S., Carr, J., Clayton, S., Dahm, C.N., Follstad Shah, J., & Sudduth, E. (2005). Standards for ecologically successful river restoration. Journal of Applied Ecology, 42(2), 208–217.

Paparisto, A., Halimi, E., Keçi, E., Hamzaraj, E., & Laknori, O. (2009). Using insects as bio-indicators to assess water quality of Albanian rivers. Journal of International Environmental Application & Science, 4(3), 246-252.

Pearson, R. G., & Boyero, L. (2009). Gradients in regional diversity of freshwater taxa. Journal of the North American Benthological Society, 28(2), 504–514.

Petersen, I., Masters, Z., Hildrew, A. G., & Ormerod, S. J. (2004). Dispersal of adult aquatic insects in catchments of differing land use. Journal of Applied Ecology, 41, 934–950.

Petersen, I., Winterbottom, J. H., Orton, S., Friberg, N., Hildrew, A. G., Spiers, D. C., & Gurney, W. S. C. (1999). Emergence and lateral dispersal of adult Plecoptera and Trichoptera from Broadstone Stream, U.K. Freshwater Biology, 42, 401–416.

Prendergast, J. R., Quinn, R. M., Lawton, J. H., Eversham, B. C., & Gibbons, D. W. (1993). Rare species, the coincidence of diversity hotspots and conservation strategies. Nature, 365(6444), 335.

Pressey, R. L., Cowling, R. M., & Rouget, M. (2003). Formulating conservation targets for biodiversity pattern and process in the Cape Floristic Region, South Africa. Biological Conservation, 112(1–2), 99–127.

Pringle, C. M. (2000). River conservation in tropical versus temperate latitudes. In: Boon, P. J., Davies, B. R., & Petts, G. E. (eds) Global Perspectives on River Conservation: Science, Policy and Practice. John Wiley & Sons, Chichester, UK, pp. 371–384.

QGIS Development Team. (2014). QGIS Geographic Information System. Open Source Geospatial Foundation Project. http://qgis.osgeo.org. Qgisorg. https://doi.org/http://www.qgis.org/

R Core Team. (2018). R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna.

Ricketts, T. H., Dinerstein, E., Olson, D. M., & Loucks, C. (1999). Who's where in North America? Patterns of species richness and the utility of indicator taxa for conservation. BioScience, 49(5), 369-381.

Roberge, J. M., & Angelstam, P. (2004). Usefulness of the umbrella species concept as a conservation tool. Conservation Biology, 18(1), 76–85.

Robson, B. J., Chester, E. T., Mitchell, B. D., & Matthews, T. G. (2013). Disturbance and the role of refuges in Mediterranean climate streams. Hydrobiologia, 719(1), 77–91.

Rubinoff, D. (2001). Evaluating the California gnatcatcher as an umbrella species for conservation of southern California coastal sage scrub. Conservation Biology, 15(5), 1374-1383.

Russi, D., ten Brink, P., Farmer, A., Badura, T., Coates, D., Förster, J., Kumar, R., & Davidson, N. (2013). The economics of ecosystems and biodiversity for water and wetlands. IEEP, London and Brussels.

Samways, M. J. (2008). Dragonflies as focal organisms in contemporary conservation biology. In: Córdoba-Aguilar, A. (ed.) Dragonflies and Damselflies: Model Organisms for Ecological and Evolutionary Research. Oxford University Press, Oxford, UK, pp. 97-108.

Samways, M.J., & Grant, P.B.C. (2007). Honing Red List assessments of lesser known taxa in biodiversity hotspots. Biodiversity and Conservation, 16, 2575–2586.

Samways, M. J., & Sharratt, N. J. (2010). Recovery of endemic dragonflies after removal of invasive alien trees. Conservation Biology, 24(1), 267–277.

Samways, M. J., & Simaika, J. P. (2016). Manual of freshwater assessment for South Africa: Dragonfly biotic index. South African National Biodiversity Institute, Pretoria, South Africa.

Samways, M. J., Sharratt, N. J., & Simaika, J. P. (2011). Effect of alien riparian vegetation and its removal on a highly endemic river macroinvertebrate community. Biological Invasions, 13(6), 1305–1324.

Schael, D. M., & King, J. M. (2005). Western Cape river and catchment signatures. Water Research Commission, Pretoria, South Africa.

Simaika, J. P., & Samways, M. J. (2009a). An easy-to-use index of ecological integrity for prioritizing freshwater sites and for assessing habitat quality. Biodiversity and Conservation, 18(5), 1171–1185.

Simaika, J. P., & Samways, M. J. (2009b). Reserve selection using Red Listed taxa in three global biodiversity hotspots: Dragonflies in South Africa. Biological Conservation, 142(3), 638–651.

Simaika, J. P., & Samways, M. J. (2012). Using dragonflies to monitor and prioritize lotic systems: A South African perspective. Organisms Diversity and Evolution, 12(3), 251–259.

Simaika, J. P., Samways, M. J., & Frenzel, P. P. (2016). Artificial ponds increase local dragonfly diversity in a global biodiversity hotspot. Biodiversity and Conservation, 25(10), 1921–1935.

Stein, B. A., Kutner, L. S., & Adams, J. S. (eds.). (2000). Precious heritage: the status of biodiversity in the United States. Oxford University Press on Demand, Oxford, UK.

Stewart, D. A. B., & Samways, M. J. (2008). Conserving dragonfly (Odonata) assemblages relative to river dynamics in an African savanna game reserve. Conservation Biology, 12(3), 683–692.

Strayer, D. L., & Dudgeon, D. (2010). Freshwater biodiversity conservation: recent progress and future challenges. Journal of the North American Benthological Society, 29(1), 344–358.

Suh, A. N., & Samways, M. J. (2005). Significance of temporal changes when designing a reservoir for conservation of dragonfly diversity. Biodiversity and Conservation, 14(1), 165–178.

Suhling, F., Sahlén, G., Gorb, S., Kalkman, V. J., Dijkstra, K.-D. B., & van Tol, J. (2015). Order Odonata. In: Thorp, J., & Rogers, D.C. (eds.) Ecology and General Biology: Thorp and Covich's Freshwater Invertebrates. Academic Press, Waltham, MA, USA, pp. 893–932.

Switzer, P. V. (1997). Factors affecting site fidelity in a territorial animal, *Perithemis tenera*. Animal Behaviour, 53(4), 865–877.

Valente-Neto, F., de Oliveira Roque, F., Rodrigues, M. E., Juen, L., & Swan, C. M. (2016). Towards a practical use of Neotropical odonates as bioindicators: Testing congruence across taxonomic resolution and life stages. Ecological Indicators, 61, 952–959.

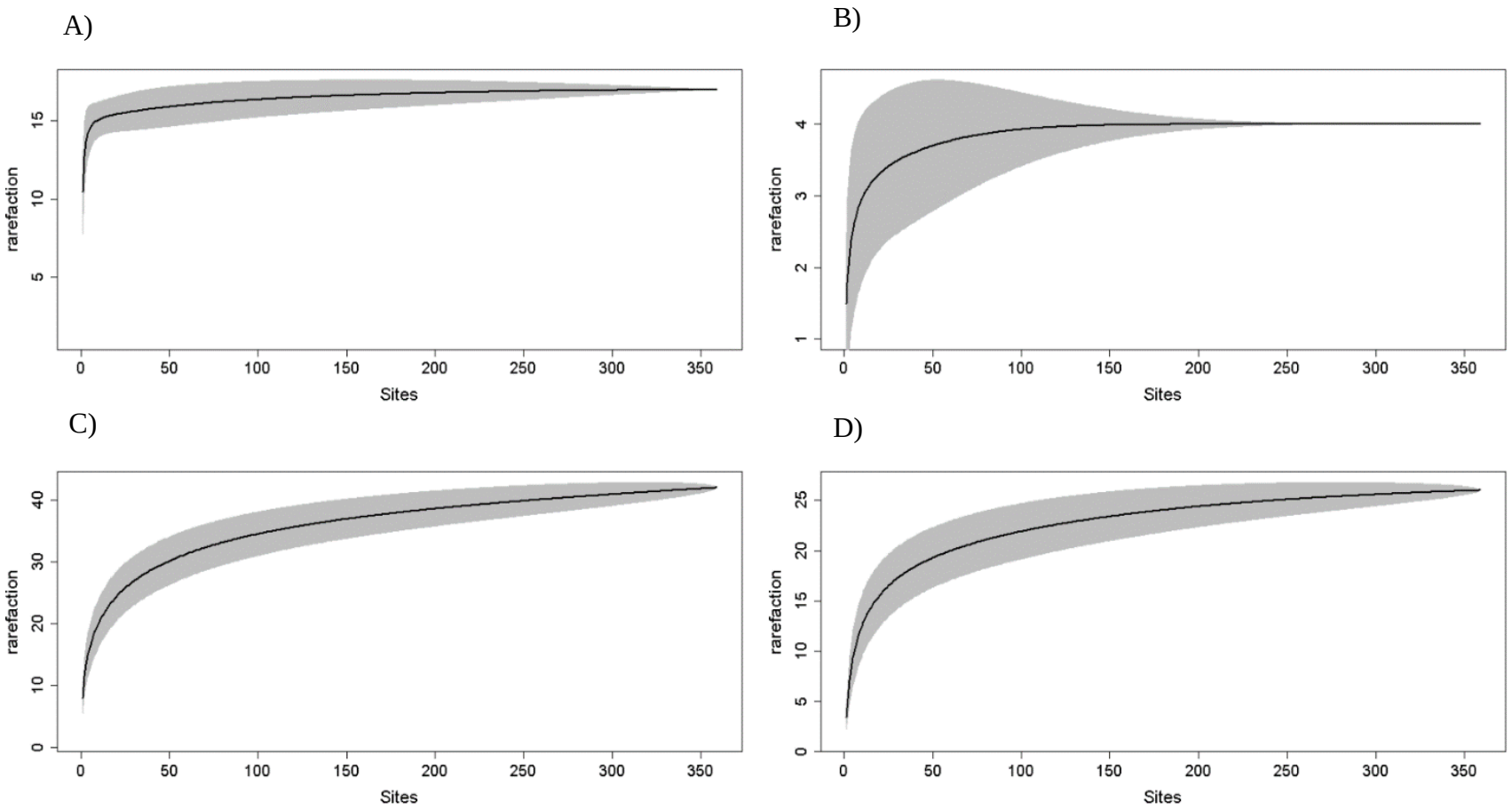
Vörösmarty, C. J., Douglas, E. M., Green, P. A., & Revenga, C. (2005). Geospatial indicators of emerging water stress: An application to Africa. Ambio, 34(3), 230–236.

Vörösmarty, C. J., McIntyre, P. B., Gessner, M. O., Dudgeon, D., Prusevich, A., Green, P., Glidden, S., Bunn, S. E., Sullivan, C. A., Liermann, C. R., & Davies, P. M. (2010). Global threats to human water security and river biodiversity. Nature, 467(7315), 555–561.

Wacholder, S., & Weinberg, C. R. (1982). Paired versus two-sample design for a clinical trial of treatments with dichotomous outcome: power considerations. Biometrics, 38(3), 801–12.

Wishart, M. J., & Day, J. A. (2002). Endemism in the freshwater fauna of the south-western Cape, South Africa. Limnologie, 28(4), 1–5.

3.7 Appendices

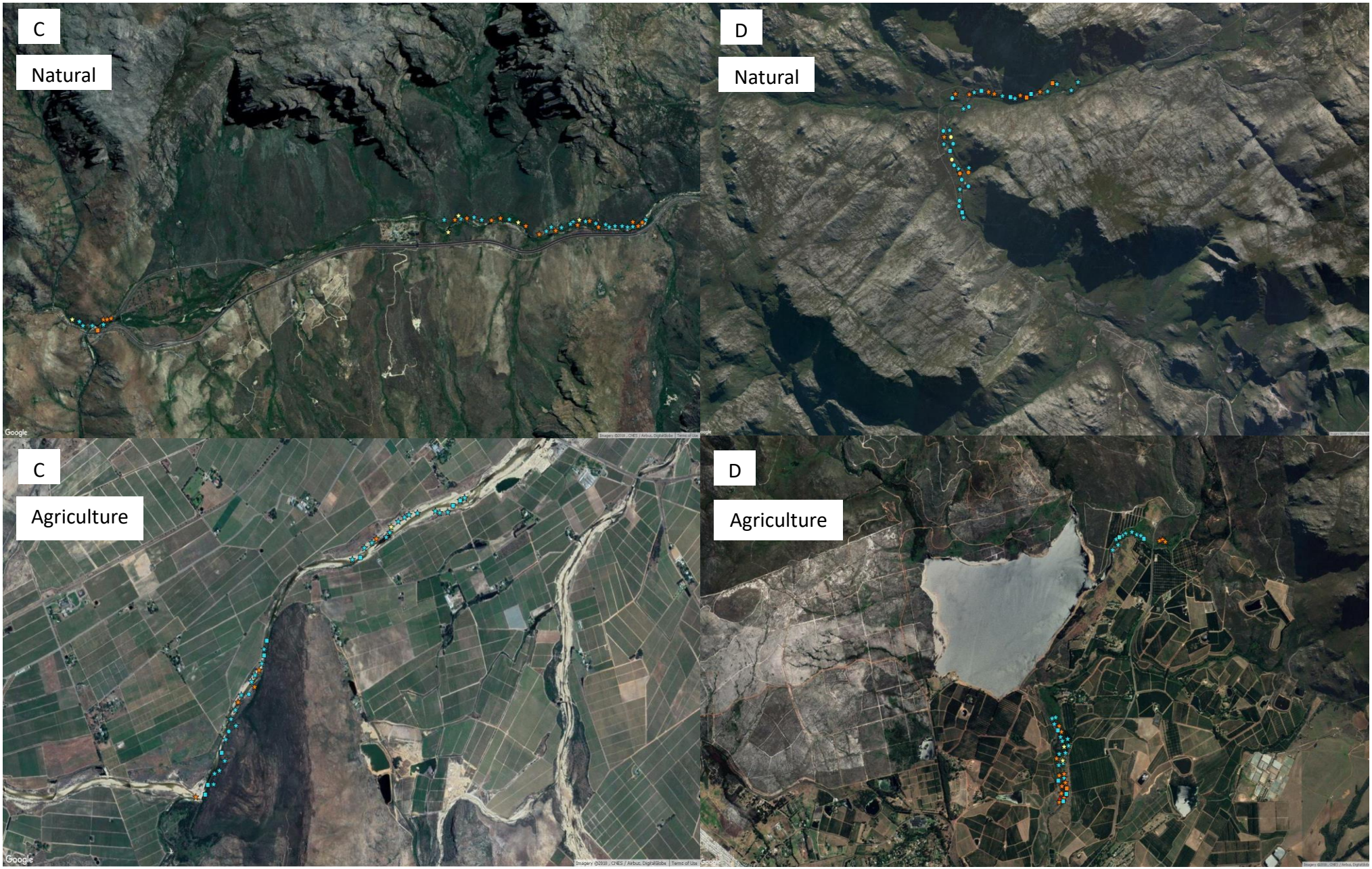


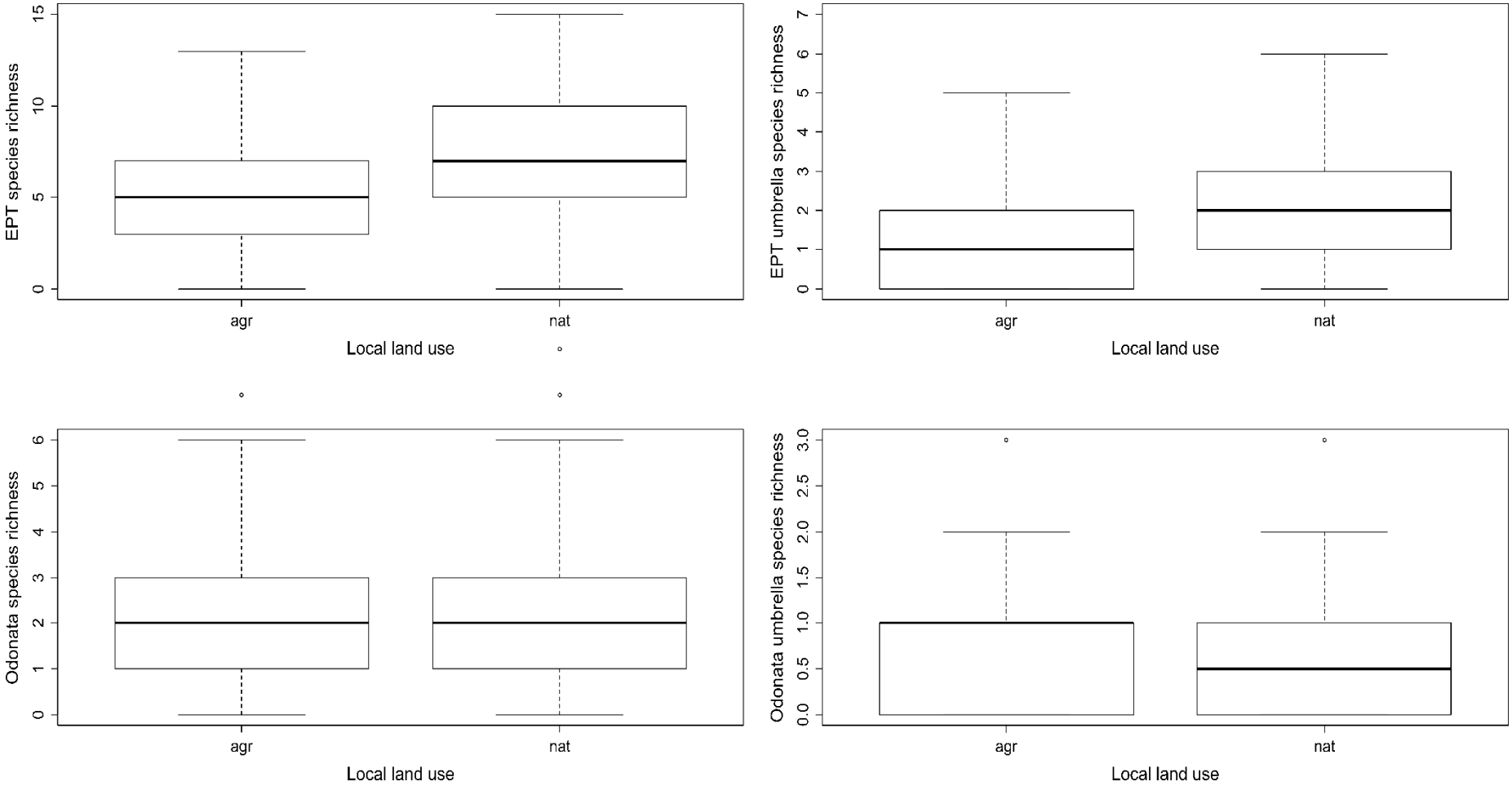
Appendix 3.A Rarefied species accumulation curves for the rivers combined for A) Ephemeroptera, B) Plecoptera, C) Trichoptera, and D) Odonata.

Appendix 3.B Odonata species names and corresponding codes.

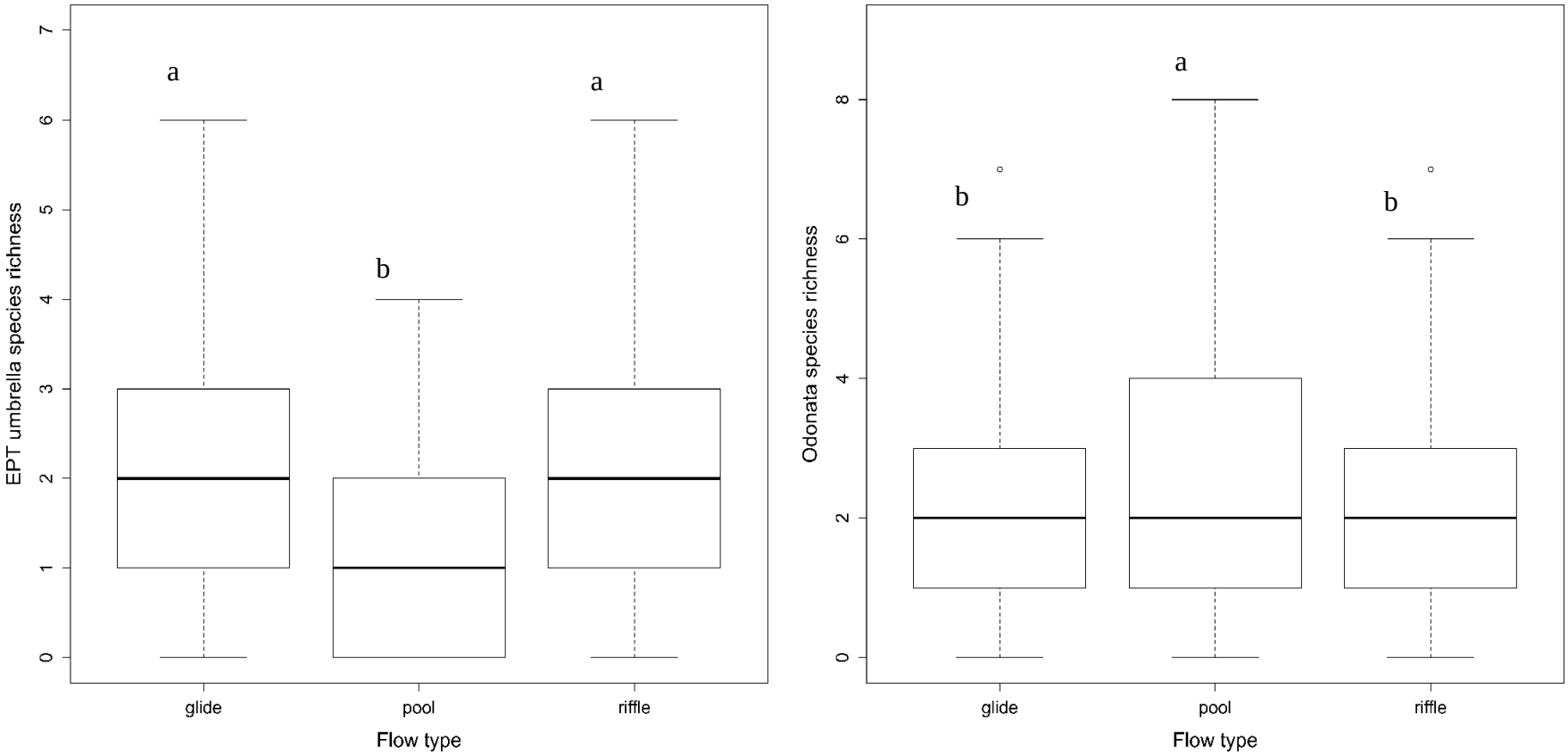
Scientific name	Species code
Suborder ZYGOPTERA	
Family CHLOROCYPHIDAE	
Platycypha fitzsimonsi (Pinhey 1950)	Pfi
Family SYNLESTIDAE	
Chlorolestes conspicuous (Hage in Selys 1862)	CC
Chlorolestes umbratus (Selys 1862)	CU
Family PROTONURIDAE	
Elattoneura glauca (Selys 1862)	EG
Elattoneura frenulata (Hage in Selys 1862)	EF
Family PLATYCNEMIDIDAE	
Allocnemis leucosticta (Selys 1863)	AL
Family COENAGRIONIDAE	
Ceriagrion glabrum (Burmeister 1839)	CG
Pseudagrion draconis (Barnard 1937)	PD
Pseudagrion furcigerum (Rambur 1842)	PF
Pseudagrion kerteni (Gersaecker 1869)	PK
Suborder ANISOPTERA	
Family AESHNIDAE	
Zosteraeschna minuscula (McLachlan 1896)	ZM
Pinheyschna subpupillata (McLachlan 1896)	PS
Anax imperator (Rambur 1842)	AI
Anax speratus (Hagen 1867)	AS
Family GOMPHIDAE	
Ceratogomphus pictus (Selys 1854)	CP
Paragomphus cognatus (Rambur 1842)	PC
Family CORDULIIDAE	
Syncordulia gracilis (Burmeister 1839)	SG
Family LIBELLULIDAE	
Orthetrum julia capicola (Kimmins 1957)	OJ
Crocothemis erythraea (Brullé 1832)	CE
Crocothemis sanguinolenta (Burmeister 1839)	CS
Trithemis arteriosa (Burmeister 1839)	TA
Trithemis dorsalis/furva (Rambur 1842/Karsch 1899)	TD
Zygonyx natalensis (Martin 1900)	ZN
Pantala flavescens (Fabricius 1798)	Pfl
Tramea limbata (Desjardins 1832)	TL

Appendix 3.C (Following page). Maps of natural and agricultural sections of the A) Eerste, B) Lourens, C) Molenaars and D) Palmiet Rivers. Colours of points indicate different numbers of sites considered (effort) where the highest number of surrogate species occurred. 72 sites (20% effort) = yellow, 180 sites (50% effort) = orange and 288 sites (80 % effort) = blue. Different shapes indicate the different groups of surrogate species, Stars = sites shared between EPT and Odonata surrogates, squares = sites selected by EPT surrogates only, circles = sites selected by Odonata surrogates only. Red triangles = sites not selected by any surrogate species. Maps extracted from Google Earth.



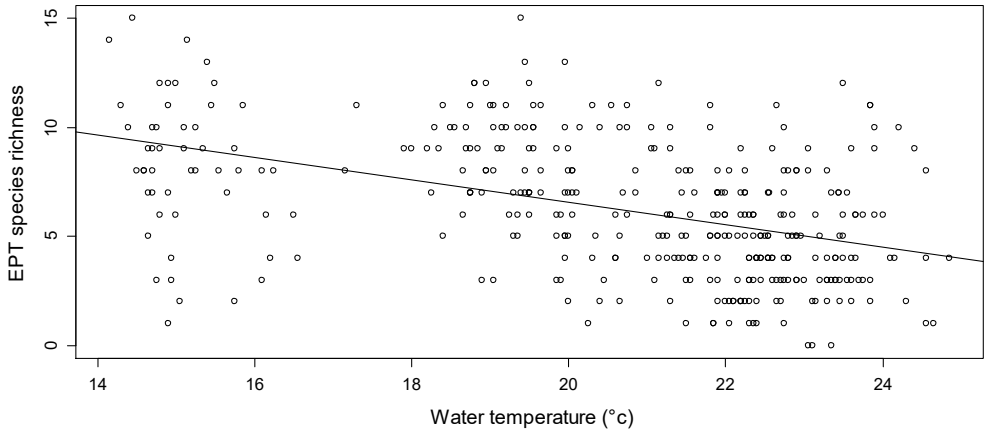


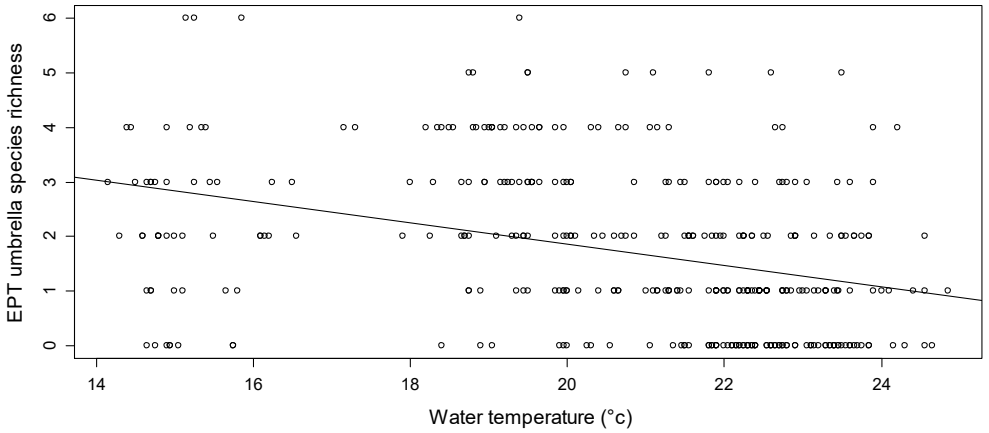
Appendix 3.D Species richness between local land use types. Agr = agriculture, nat = natural. EPT = Ephemeroptera, Plecoptera and Trichoptera combined.

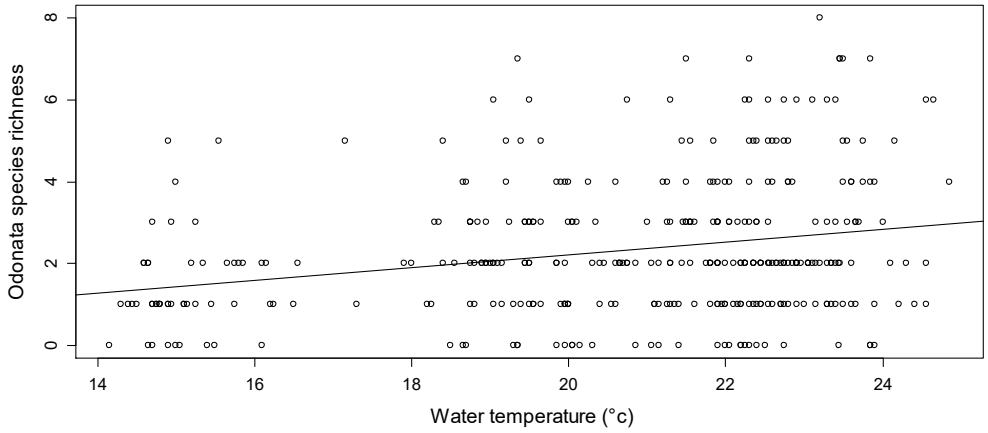


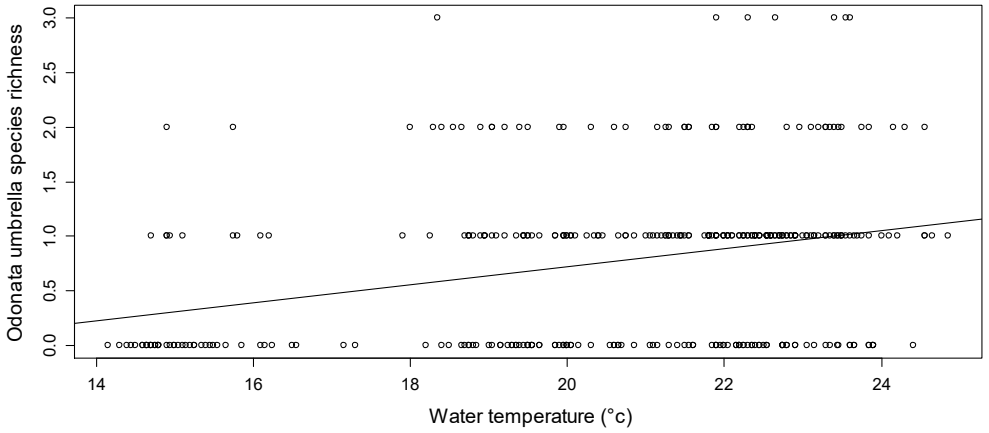
Appendix 3.E Species richness between flow types. Small letters denote significant differences. EPT = Ephemeroptera, Plecoptera and Trichoptera combined.

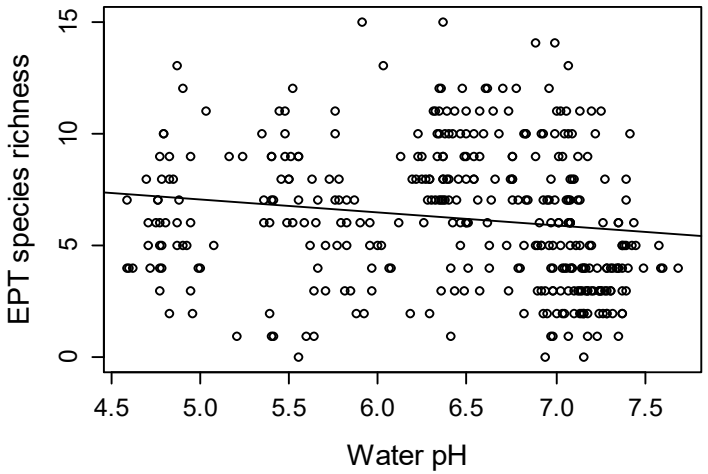
Appendix 3.F Water temperature and pH effects on species richness for Ephemeroptera/ Plecoptera/ Trichoptera and Odonata.

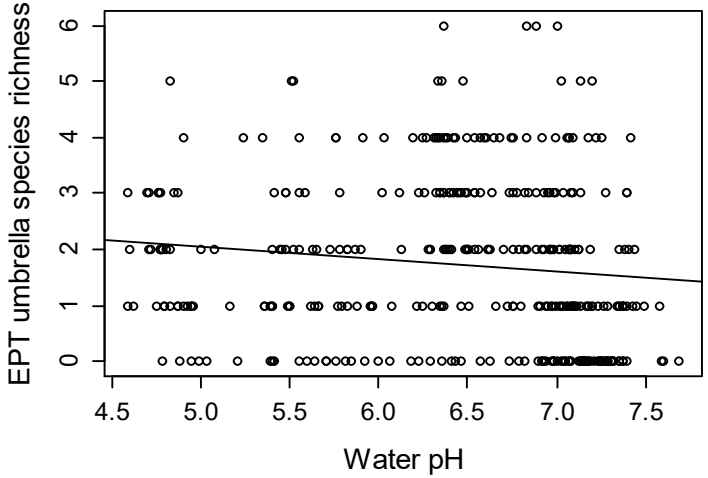


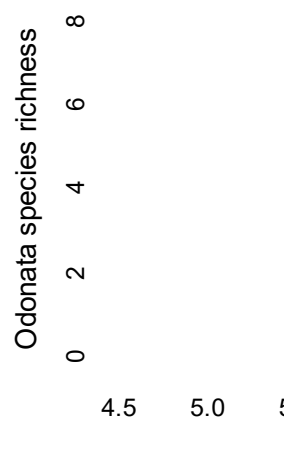


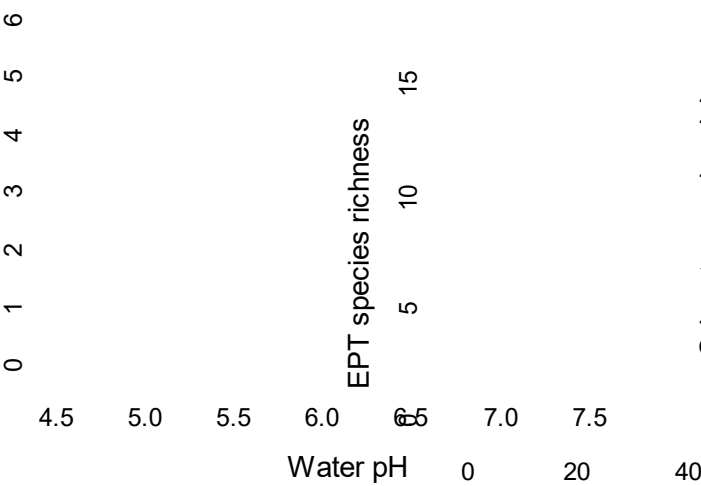


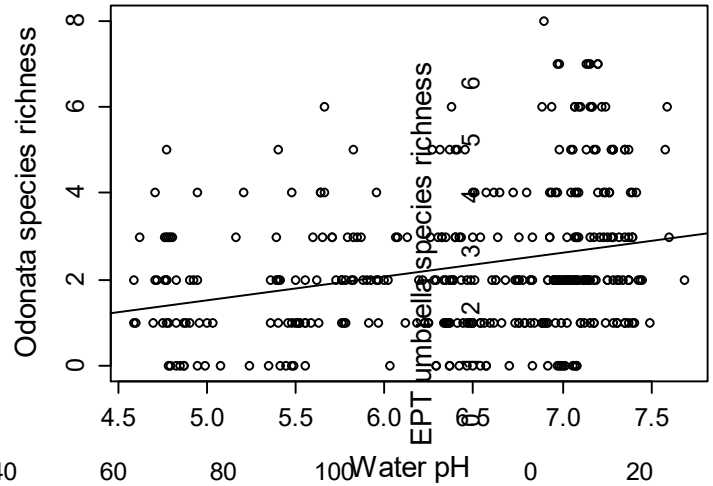


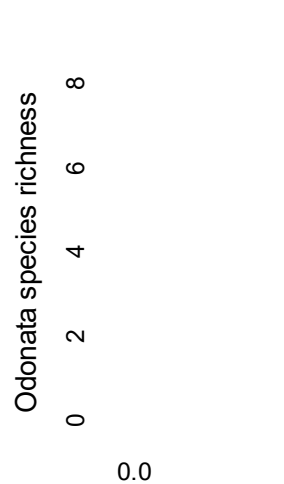


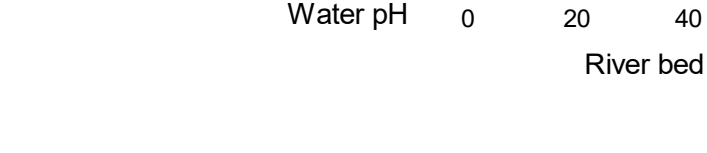


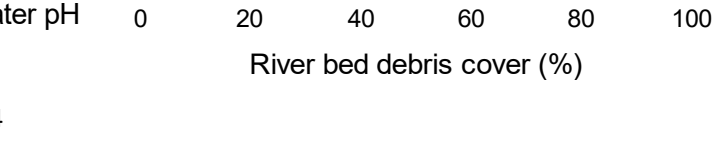












Chapter 4: Well-designed conservation ecological networks maintain river ecosystem integrity and dragonfly diversity in forestry plantations

Abstract

Landscape transformation is detrimental to ecosystem health. It results in habitat loss and fragmentation, two of the greatest causes of biotic extinctions. Timber plantations are a major threat to grassland ecosystems in southern Africa. However, incorporating large, landscape-scale ecological networks (ENs) into plantation landscapes reduces the negative effects of fragmentation by connecting natural areas, using conservation corridors of natural grassland. The benefits of ENs are not limited to only terrestrial biodiversity, as they also aim to conserve freshwater biota. Dragonflies are highly sensitive to habitat condition, and are well-known bioindicators. Here, I compare EN sites to those in nearby protected areas (PAs) to determine the effectiveness of ENs for conserving dragonfly diversity and maintaining river ecosystem integrity. I also investigate the effect of corridor width on dragonfly assemblages in ENs. Lastly, I explore which environmental variables are important drivers of dragonfly diversity and which species traits are being selected for within the ENs and PAs. Along river edges, adult male dragonflies were recorded, with 27 environmental variables at 55 EN and 55 PA sites. Dragonfly diversity and the measured environmental variables did not differ between the EN and PA sites. Dragonfly abundance and species richness were positively correlated with EN corridor width. In-water variables, as well as elevation (m a.s.l.), were the most important drivers of dragonfly assemblages for both ENs and PAs. The EN as a mitigation approach in the context of plantation forestry is shown here to be highly effective for lotic systems, and should continue to be implemented as part of an overall environmental protection plan in transformed landscapes.

4.1 Introduction

Landscape transformation is a key threat to ecosystem integrity, and the negative impacts associated with it extend far beyond the boundaries of the transformed land (Vitousek et al., 1997). Sensitive ecosystems, such as rivers, are the most severely affected by transformation (Burkey, 1995; Kinvig & Samways, 2000). For biota, anthropogenic development causes habitat loss and fragmentation, which are the two greatest causes of biotic extinctions (Fahrig, 1997), and conservation efforts are often aimed at reducing these impacts. The two issues that arise from fragmentation are a decrease in habitat size and the division of any remaining habitat into isolated patches (Wilcove et al., 1986; Noss, 1987). The theory of island biogeography explains the importance of fragment size and isolation (MacArthur & Wilson, 1967; Simberloff & Abele, 1976). Although the concepts behind the theory remain relevant, other studies have highlighted the significance of metapopulation dynamics in terrestrial environments (Howe et al., 1991; Fahrig & Merriam, 1994). Isolated habitat patches may not be able to maintain species populations unless there is ongoing exchange of genetic material from other populations (Noss, 1987).

To prevent species extinctions and to ensure the long-term maintenance of biodiversity, it is necessary to connect patches of remaining natural habitat to each other (Fahrig & Merriam, 1994; Samways, 2007). It is in this context that conservation corridors of natural or restored vegetation have become a favoured approach in production landscapes (MacDonald, 2003). A conservation corridor can facilitate animal movement, but can also function as a habitat, provided the required resources are available. Globally, corridors have become a well-established practice in urban (Rosenberg et al., 1997; Bennett, 1999; Jongman & Pungetti, 2004; Samways, 2007), agricultural (Rodenhouse et al., 1992; Fedorowick, 1993; Cowling et al., 2003; LaRue & Nielsen, 2008), and plantation landscapes (Bennett, 1999; Lemckert et al., 2005; Basiron, 2007; Samways et al., 2010).

For timber plantations in the KwaZulu-Natal Province of South Africa, the collective landscape linkages and adjoining natural habitat patches are referred to as ecological networks (ENs) (Bazelet & Samways, 2011; Hansen et al., 2018). These ENs comprise mostly linear patches of remnant land (often connecting protected areas (PAs) outside of ENs) connected together using corridors and stepping-stone habitats (Pryke et al., 2015; Joubert et al., 2017; Hansen et al., 2018). For terrestrial biota, they increase dispersal of organisms, including small mammals, butterflies (Haddad, 1999; Pryke & Samways, 2001) and bird-dispersed plants (Haddad et al.,

2003). They also effectively maintain the diversity of many arthropod taxa (Pryke & Samways, 2012; Pryke et al., 2015).

There is an urgent need for further research on conservation corridors to define the best approaches for improving functional connectivity across the transformed landscape. Noss (1987) and Simberloff and Cox (1987) emphasized that research findings and recommendations on corridor design should not be applied universally, and that corridors need to be individually evaluated. The extent to which a corridor may alleviate the effects of fragmentation depends upon numerous considerations. The major constituents are the region, transformation type and intensity, size of the corridor, the habitat structure (environmental variables) within the corridor and the autecology of the local biota (Forman & Godron, 1986; Noss & Harris, 1986; Noss, 1987).

Here, I use the above-mentioned criteria to determine the success of ENs for conserving river health and dragonfly diversity in plantation landscapes of the KwaZulu-Natal Province, South Africa. Although small rivers and streams are commonly included in corridors, the importance of their functional attributes are often overlooked in research on terrestrial corridors. Rivers are highly sensitive systems, and are affected by all landscapes through which they flow (Jongman, 2002; Stein et al., 2002). Their continuum nature means that any anthropogenic impact that occurs at a point is also carried down the river (Jongman, 2002). For example, clear-felled plantations increase run-off into rivers through removal of topsoil along with its valuable components, like minerals, which are then transported down the river (Forman, 1995). These impacts influence numerous river dynamics, such as those related to flow rate and water chemistry, which in time, impacts biodiversity (Forman, 1995; Kinvig & Samways, 2000). In some rivers, biodiversity losses are estimated to be five times greater than in degraded terrestrial ecosystems (Dudgeon et al., 2006; Simaika & Samways, 2009). For KwaZulu-Natal plantations, it was recommended that ENs include rivers with unplanted areas > 30 m to either side. This is to buffer the effects that plantations have on river systems and their biota (Kinvig & Samways, 2000; Samways & Pryke, 2016).

Odonata (encompassing true dragonflies and damselflies but hereafter will be collectively referred to as dragonflies) is the focal taxon here. They are an order of well-studied, semi-aquatic insects that are also recognized as umbrella species, whose conservation results in the protection of other co-occurring species (Samways & Simaika, 2016; Chapter 2). They are utilized as sensitive indicators of habitat quality and landscape disturbance. This potential

results in their extensive use as valuable components of various indices (Samways, 2008; Samways & Simaika, 2016). One example is the Dragonfly Biotic Index (DBI), which uses the presence of dragonfly species as a measure of freshwater condition. The index has successfully monitored restoration projects and identified sites requiring conservation management (Samways & Taylor, 2004; Simaika & Samways, 2011; 2012; Harabiš & Dolný, 2012). Kietzka et al. (2015) found that only natural environmental variables significantly influenced DBI scores in wide, high quality ENs in KwaZulu-Natal, with little affect from anthropogenic disturbance. However, no studies have attempted to assess the complementarity between lotic freshwater conditions across a range of EN corridor widths in comparison with natural grasslands in neighbouring PAs. The main aim here is to determine the effectiveness of ENs at mitigating the influence of timber production landscapes on river health, in particular, in terms of dragonfly diversity. To achieve this, I compare dragonfly diversity between rivers within timber ENs to nearby untransformed rivers in PAs surrounded by natural grassland. Furthermore, I test whether wide corridors support greater dragonfly diversity than narrow corridors, and so specifically test the influence of corridor width on dragonfly diversity. I also determine the environmental drivers of dragonfly diversity within ENs and PAs. To gain a more mechanistic understanding of environmental influences on dragonfly assemblages in these systems, I test which dragonfly functional and life-history traits are being selected for by different environmental variables within ENs and PAs. This will help determine which factors are important for conservation managers to consider in planning and managing these landscapes. Complementary effects between natural and EN sites has been found under subtropical conditions, where ponds in ENs and neighbouring PAs showed equivalence in dragonfly assemblages (Pryke et al., 2015). However, I am not expecting such a distinct result, as rivers are more responsive to the condition of the catchment as a whole, compared to ponds. Nevertheless, I hypothesize that the wide corridors will retain the highest diversity, as they would be expected to have higher habitat heterogeneity, which is a major driver of species richness (Kietzka et al., 2015). Furthermore, wider corridors would presumably be more buffered from the effects of plantations compared to narrower corridors.

4.2 Materials and methods

4.2.1 Study area

Study sites were established on various commercial plantations, owned by the Mondi Group, across the KwaZulu-Natal Midlands, South Africa (Fig. 4.1). The plantations comprise *Eucalyptus, Acacia* and *Pinus* species. Sites included as much variation as possible between corridors in terms of corridor width, invasive plant species and cover, vegetation height, shade and river size. Reference sites comprised rivers within PAs or those surrounded by large areas of natural vegetation (Fig. 4.1). The landscape typically comprises small hills, rocky outcrops and clay soils (Pryke & Samways, 2015). Natural vegetation in the region is dominated by the threatened Midlands Mistbelt Grassland and Drakensberg Foothill Moist Grassland (Muchina & Rutherford, 2006). Temperatures typically range between -2°C and 38°C, with a high annual rainfall between 800-1280 mm, received mostly in summer (Lipsey & Hockey, 2010). A total of 55 PA and 55 EN sites were chosen and each comprised a 100 m transect of river parallel to the river bank, and 5 m to either side of it. Sites were separated by a minimum distance of 1.5 km. Each site was visited twice during the summer months, from January 2018 to March 2018.

4.2.2 Dragonfly sampling

Adult, male dragonflies were surveyed for an hour per visit per site by two observers. Species and their abundances were recorded by observation using close-focus binoculars. Hand held insect nets were used to capture dragonfly individuals for voucher specimens, and when individuals were difficult to identify in flight. Observations were conducted during the summer months at peak periods of activity, generally from 9:00 until 15:00. Surveys were avoided when weather conditions were overcast, rainy or windy. Specimens were identified and DBI scores obtained using the protocol of Samways and Simaika (2016).

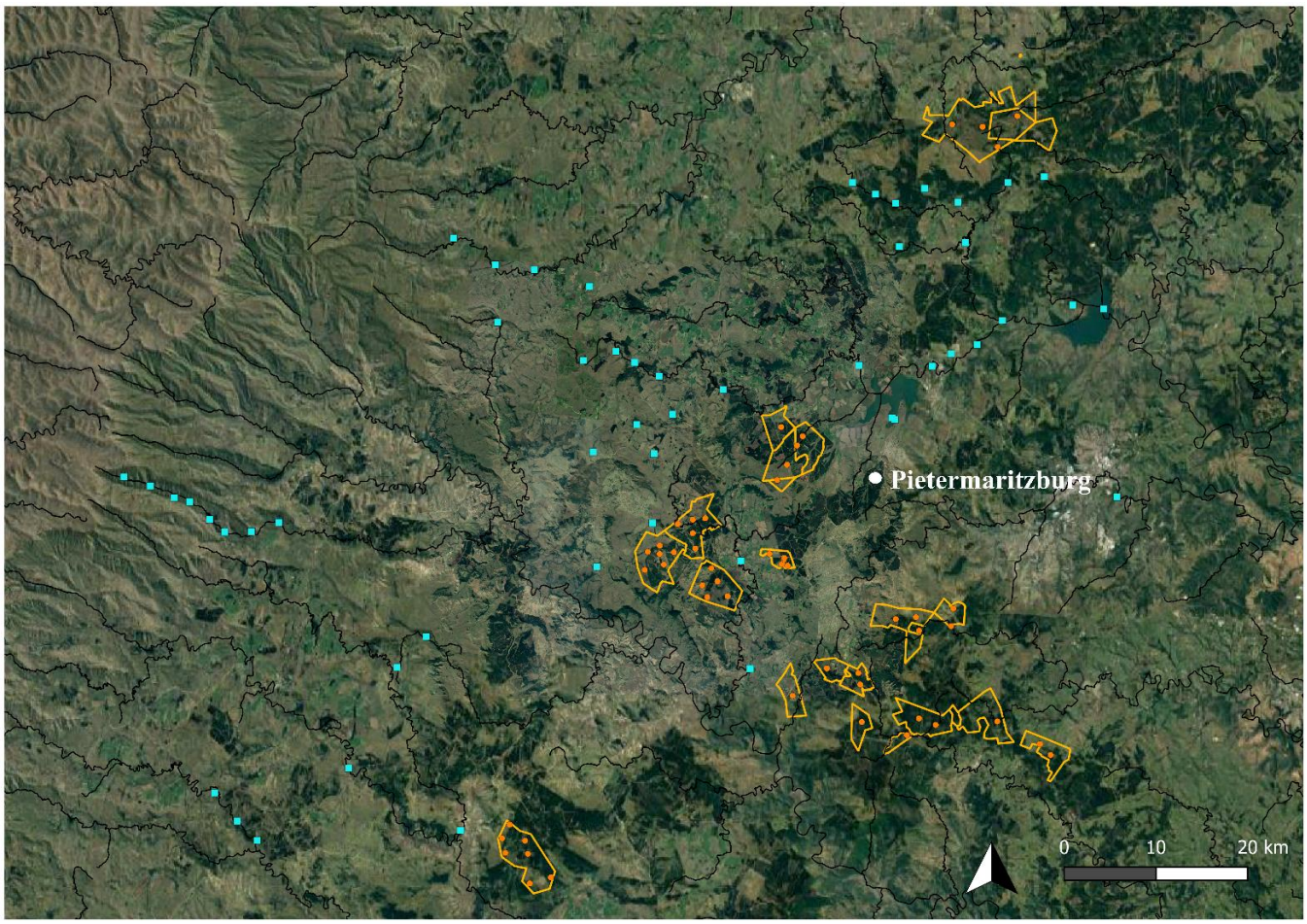


Fig. 4.1 Map of sites in KwaZulu-Natal. Blue squares represent natural sites and orange circles represent plantation sites situated within the plantation boundaries outlined in orange. EN data were obtained from Mondi Group and the map layer was exported from Google Earth.

4.2.3 Dragonfly Biotic Index scores

The DBI score per site was calculated as the average of all the adult male species scores recorded at that site. (Samways, 2008; Simaika & Samways, 2009; Kietzka et al., 2015; Samways & Simaika, 2016). Each species has its own particular DBI score (zero to nine) based on its geographical range, threat status according to the IUCN Red List, and its sensitivity to habitat disturbance. Species with a score of zero are geographically widespread, have a Least Concern threat status, and are tolerant of anthropogenic habitat disturbances. At the other end of the spectrum, species with a score of nine have small geographical ranges (are usually a national endemic), are Endangered or Critically Endangered, and extremely sensitive to habitat disturbance (Samways & Simaika, 2016). The average DBI score of all the species at a site

defines that site in terms of its condition. A low site score would consist mostly of an assemblage of widespread habitat generalists, and a high site score an assemblage containing rare, threatened, specialists.

4.2.4 Environmental variables

A total of 27 environmental variables were recorded at each site (Appendix A). Land use (whether the sites were in an EN or PA) was recorded as a categorical variable. The river width and depth were measured with a tape measure. When river sites were too deep to measure they were recorded to have >30 m depth. Elevation and geographical position were determined using a handheld GPS. Percentage shade (at midday), cattle activity, rocks, sand and detritus covering each site were estimated as percentage cover by both observers at the time of data collection and averaged. Similarly, each site was divided between the flow rates of pool, riffle and glide. A 'Clarity tube' (designed by GroundTruth, a South African company based in Hilton) was used to measure water turbidity at each site. Measuring of in-water variables (water temperature, dissolved oxygen, conductivity and pH) was done with a handheld dissolved oxygen meter (Model: HI9146; Make: HANNA Instruments) and a Combo Meter (Model: HI98129; Make: HANNA Instruments). Vegetation data comprised the average height of the riparian vegetation, the percentage cover of emergent aquatic macrophytes in the river and along its edges and the percentage cover of the three most abundant alien plant species (*Rubus cuneifolius, Solanum mauritianum* and *Cirsium vulgare*) were each recorded separately. The cover of any additional alien plant species were added together as a separate group. A DEM model was created using Google Earth, and this was used to obtain corridor width, slope, aspect and percentage of transformed area within a 1 km and 5 km radius of each site.

4.2.5 Dragonfly species traits

Species traits were used to analyse the effect of environmental variables on species life-history strategies. Traits used here were: sensitivity to habitat change according to the DBI, whether the species spent the majority of its diurnal time perching or flying, the water body type (WBT) with which the species is associated, preference for lentic or lotic habitat type, whether presence of rocks was an important habitat requirement, tolerance to shade, family, suborder, the maximum and minimum elevations at which a species was recorded, the length of flight

season, and average hindwing and body length of the species. All trait information was obtained from Samways and Simaika (2016).

4.2.6 Statistical analyses

In R version 3.5.0 (R Core Team, 2016), in the *vegan* package (Oksanen et al., 2008), the function *specpool* was used to determine whether sampling effort was sufficient. The function estimates the number of unseen species that belong to the species pool, and assumes that the number of unseen species is related to the number of rare species or those observed only once or twice. The non-parametric species estimators Chao2 and ICE were calculated, and a site based rarefaction curve was produced.

Moran's I autocorrelation index showed sites were spatially autocorrelated, and therefore a random spatial variable was used in all analyses. This variable was based on the division of sites location categories according to the sites geographical clusters (Fig. 4.1) (Gittleman & Kot, 1990).

Spearman's rank correlation matrices of the environmental variables were used to identify any correlated variables. When two variables were strongly correlated ($r > 0.5$), one variable, the most ecologically intuitive variable based on knowledge of dragonflies responses to environmental variables, was retained, while the other was removed.

Summary statistics were calculated in R to analyse how similar or different the environmental variables were between EN and PA sites.

Shapiro-Wilk normality tests were run on dragonfly abundance, species richness and DBI scores (Royston, 1995). Dragonfly abundance and species richness data were not normally distributed, but DBI scores were. All generalized linear mixed-effect models (GLMMs) and linear mixed-effect models (LMMs) were performed in R (R Core Team, 2016) using the lme4 package and had location as the random variable (Bates et al., 2014). For all abundance and species richness models, GLMMs were created, and for the normally distributed DBI scores, LMMs were used. All GLMMs were fitted by a Laplace approximation with Poisson error distribution and log-link function (Bolker et al., 2009), and LMMs with Gaussian distribution (Oberg & Mahoney, 2007).

To answer the first objective, dragonfly abundance, species richness and DBI scores were analysed to determine whether they differed between the EN and PA sites. For the second objective, only plantation EN sites were used, and models were created to determine the influence of corridor width on the same three assemblage measures. For the final objective, the *glmulti* package (Calcagno & de Mazancourt, 2010) was used to run automated model selection procedures (AMS) with the main effects based on second-order Akaike information criterion (AICc). These models were run with dragonfly species richness and DBI scores as the response variable for EN and PA sites combined, and each separately. This procedure selects variables, or combinations of variables, that improve the linear model on which AMS is based. Thereafter, GLMMS were used to identify which of the selected explanatory variables from the AMS models significantly influenced dragonfly species richness and DBI scores. The significant variables were plotted as dot and whisker diagrams in the R package *dotwhisker* (Solt et al., 2018) using the *dwplot* function. As none of the models showed overdispersion, likelihood ratio-tests were used to calculate χ^2- and P- values (Bolker et al., 2009).

For assemblage composition analyses, the function manyglm in the *mvabund* package (Wang et al., 2014) was used to determine the best fit distribution for the data. Abundance data were used to apply generalised linear models (GLMs) with multivariate extensions. These were best modelled with a negative binomial distribution assuming a quadratic mean-variance. This method allows for the multiple effects at the assemblage level to be tested. Such model-based approaches have better power properties than distance-based approaches. The *manyglm* function fits a separate GLM to each species, using the given explanatory variables. It then uses resampling-based hypothesis testing, while also accounting for correlations between species, to make assemblage-level conclusions about which environmental variables are associated with the multivariate abundances (Wang et al., 2012). For the first objective, the model looked at the effect of PAs vs. ENs on community composition, and for the second objective, corridor width was included as the fixed effect. For the third objective, in order to select the most important environmental variables that affected dragonfly assemblage structure, Canonical Correspondence Analyses (CCAs) were run in CANOCO 5 (ter Braak & Šmilauer, 2012). This was done for PA and EN sites combined, and each separately. Interactive forward selection analyses (ter Braak, 1990) were used to select the best group of variables, according to the amount of variation in the dragonfly data that they explained after accounting for the variation of the other variables. Analyses were permutated 999 times to normalize distribution and allow comparisons of variables (Lepš & Šmilauer, 2003). The significant variables selected by the

CCAs were divided into three groups: natural, in-water and anthropogenic variables. Each group (up to four variables) was run separately for the assemblage analyses in R (R Core Team, 2016).

Multivariate abundance data were analysed with the *Boral* (Hui, 2016) package in R (R Core Team, 2016). This produced model-based analyses using Bayesian Markov Chain Monte Carlo estimation methods (Hui, 2016). This approach to unconstrained ordination makes use of pure latent variable models, and those that include the influence of environmental variables, which gave detailed visualization of the similarity in dragonfly assemblages between EN and PA sites.

To determine how the abundance of individual dragonfly species were influenced by their functional traits, the fourth-corner modelling approach was used (Warton, Shipley & Hastie, 2015). Separate analyses were run for EN and PA sites using the *traitglm* function in the package *mvabund* (Brown et al., 2014; Wang et al., 2014) in R (R Core Team, 2016). This method constructs a regression model for dragonfly abundance as a function of the interaction terms between environmental variables and traits (Brown et al., 2014). Data fitted a negative binomial distribution and the least absolute shrinkage and selector operator (LASSO) model-selection argument *glm1path* was used to interrogate correlated trait-predictors. This function zeroes the coefficients for any model interactions that do not lower the Bayesian information criterion (Brown et al., 2014; Rees et al., 2018). The *levelplot* function from the *lattice* package in R (R core team, 2016) was used to graphically represent the fourth-corner coefficients.

4.3 Results

4.3.1 Dragonfly species abundance and richness

A total of 1 362 dragonfly individuals (20 Anisoptera species and 17 Zygoptera species) were sampled. EN sites had 742 individuals (20 Anisoptera species and 16 Zygoptera species) and PA sites 620 individuals (18 Anisoptera species and 15 Zygoptera species) (Appendix 4.A species list). Four species, *Africallagma sapphirinum*, *Tramea basilaris*, *Crocothemis erythraea* and the rare, highly threatened, and very sensitive *Chlorolestes apricans*, were recorded only in EN sites and were absent PA sites. One species, *Pseudagrion hageni*, was only recorded at PA sites. The rarefication curve reached an asymptote, indicating that

sampling effort was sufficient to represent the species present (Appendix 4.B). Additionally, this was supported by species estimates (Sobs = 37; Chao2 = 38.18 (±1.83); ICE = 38.77).

4.3.2 The influence of ecological networks on dragonfly assemblages

For dragonfly abundance (χ^2 = 1.80; P = 0.18), species richness (χ^2 = 0.30; P = 0.59) and DBI scores (χ^2 = 0.10; P = 0.76) there were no significant differences between EN and PA sites (Fig. 2). Dragonfly species composition also did not differ (LRT = 50.48; P = 0.11). Summary statistics of the environmental variables between EN and PA sites showed them to be very similar (Appendix 4.C).

4.3.3 The influence of corridor width on dragonfly assemblages

Dragonfly abundance (χ^2 = 3.70; P < 0.05) and species richness (χ^2 = 9.86; P < 0.001) were both significantly and positively correlated with corridor width (Fig. 4.3). However, DBI scores (χ^2 = 0.10; P > 0.05) were not significantly influenced. Dragonfly assemblage composition was influenced by corridor width (LRT = 110.1; P < 0.05).

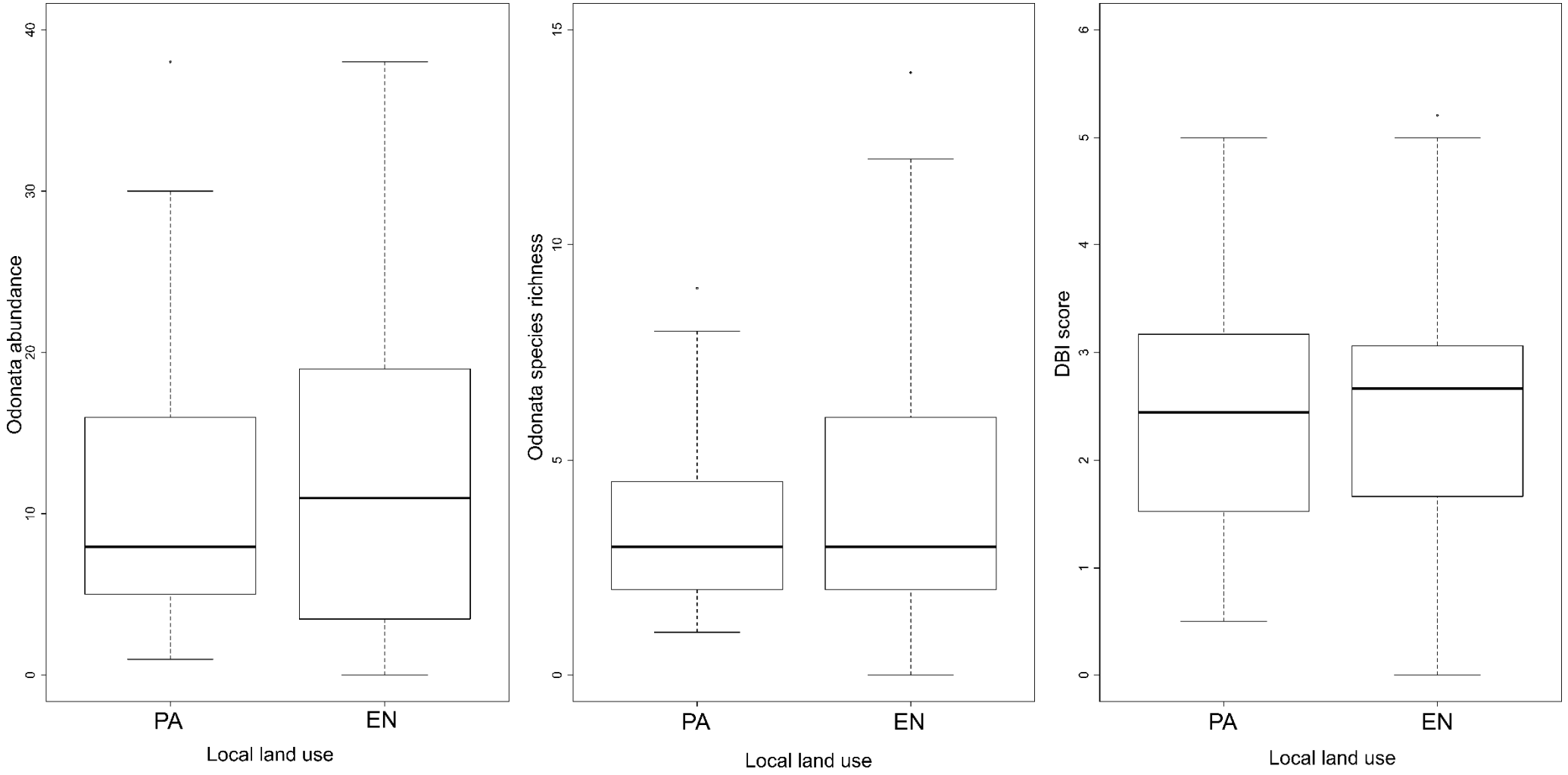


Fig. 4.2 Box plots of dragonfly abundance, species richness, and DBI scores between PA (protected area) and EN (ecological network) sites.

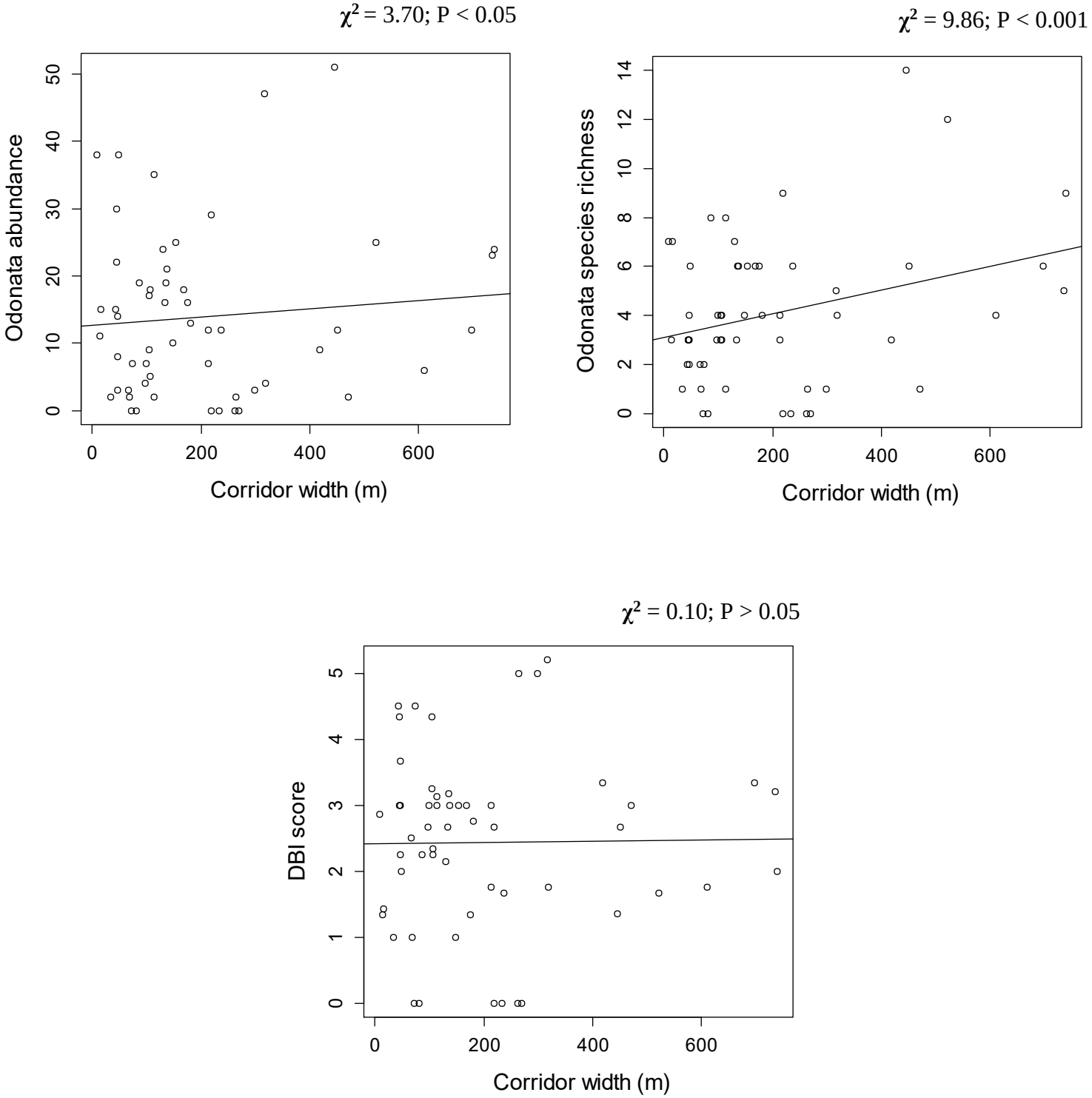


Fig. 4.3 Effect of corridor width on dragonfly abundance, species richness, and DBI scores for EN sites.

4.3.4 Variables influencing dragonfly species richness

For EN and PA sites combined, percentage of shade, invasive species and elevation, were the three most important factors, all negatively correlated with species richness (Fig. 4.4; Appendix 4.D). This was closely followed by percentage of dissolved oxygen, which was positively related to species richness. Furthermore, slope and the percentage of the alien plant *S. mauritianus* also negatively correlated with overall species richness. In contrast, water turbidity, the area of transformed land within a 5 km radius and the percentage of riffles were positively correlated. For EN sites, dissolved oxygen was the most significant variable, and along with river depth and percentage of riffles, positively correlated with species richness (Fig. 4.4; Appendix 4.D). Invasive alien plant cover and the alien *S. mauritianum*, were the second and third most influential variables and negatively influenced species richness, along with aspect. For PA sites, only elevation and shade significantly and negatively correlated with species richness (Fig.4.4; Appendix 4.D).

4.3.4 Variables influencing Dragonfly Biotic Index scores

The in-water variables, temperature and turbidity, negatively correlated with DBI scores for the sites overall (Fig. 4.4; Appendix 4.D). Water temperature also negatively influenced DBI scores in the PA sites and was the only significant variable (Fig. 4.4; Appendix 4.D). For EN sites only water conductivity significantly, negatively correlated with DBI scores (Fig. 4.4; Appendix 4.D).

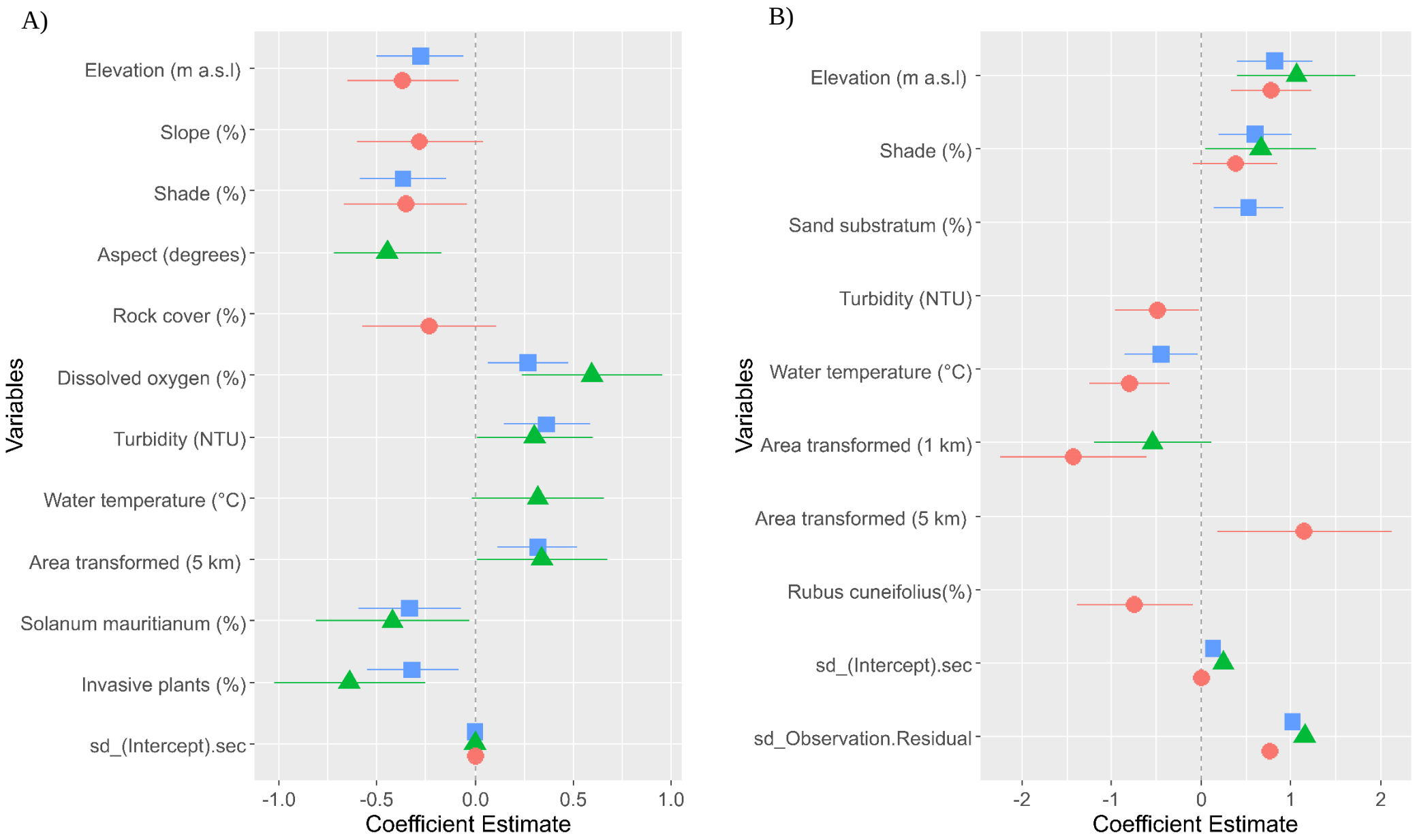


Fig. 4.4 Dot and whisker plots for A) species richness and B) DBI scores. Squares = sites combined, triangles = plantation sites only and circles = natural sites only. Dots show regression coefficient point estimates and whiskers at the confidence interval (95%).

4.3.5 Dragonfly species assemblage composition

For the EN and PA sites combined, elevation was the only significant landscape variable, with the greatest influence on the dragonfly assemblage composition (Table 4.1). This was followed by the water conductivity. The percentages of alien *C. vulgare*, a management variable, influenced dragonfly assemblages, significantly interacted with percentage of debris (Table 4.1). For EN sites, dragonfly assemblages were most significantly influenced by water parameters, namely temperature and conductivity. Of the landscape variables, elevation and the percentage rocks significantly influenced assemblage composition, while the presence of cattle was the only significant management variable (Table 4.1). For PA sites, landscape and water measures significantly influenced assemblage composition. Of the landscape variables, elevation, shade cover, slope, and percentage rock were all significant (Table 4.1). Shade cover interacted with slope and elevation, which suggests that they both have an influence on the effect of shade cover (Table 4.1). Water turbidity, conductivity, river width, and pH significantly affected the composition of the assemblage. Furthermore, turbidity and water temperature interacted at PA sites.

The pure latent variable model of local land use for dragonflies showed almost no separation between the EN and PA sites (Fig. 4.5). When environmental variables were included in the model, all of the sites grouped closer together. Overall there was still no observable separation between the EN and PA sites, and so land use did not explain the model well.

Table 4.1 Dragonfly assemblage response in the plantation and natural sites combined, and separately, using multivariate generalised linear modelling on the effects of environmental variables on species abundance data.

Variables		Deviance test statistic		
		Overall	**Plantation**	**Natural**
Natural variables	Elevation (m a.s.l)	70.66**	84.03**	101.01***
	Aspect (degrees)			
	Slope (%)			84.64***
	Shade (%)			93.18
	Sand substratum (%)			
	Rock side (%)	47.91	49.38*	53.71
	Slope (%) x Shade (%)			69.41**
	Elevation x Shade			41.22*
In-water measures				
	Water temperature (˚C)		103.66**	34.71
	Conductivity (ppt)	64.58*	59.45*	106.02**
	pH	37.95		58.12*
	Dissolved oxygen (%)	46.04		
	Turbidity (NTU)			116.03***
	Width (m)			71.63**
	Water temperature x Turbidity			52.08
Anthropogenic variables				
	Area transformed (1 km)			30.11
	Area transformed (5 km)		62.64	
	Rubus cuneifolius (%)		51.30	
	Solanum mauritianum (%)	49.43		
	Invasive plants (%)			35.51
	Corridor width (m)			
	Cattle presence (%)		61.43*	
	Cirsium vulgare (%)	47.10*		
	Debris (%)	45.81		
	C. vulgare x Debris	16.61*		

* P < 0.05, ** P < 0.01, *** P < 0.001

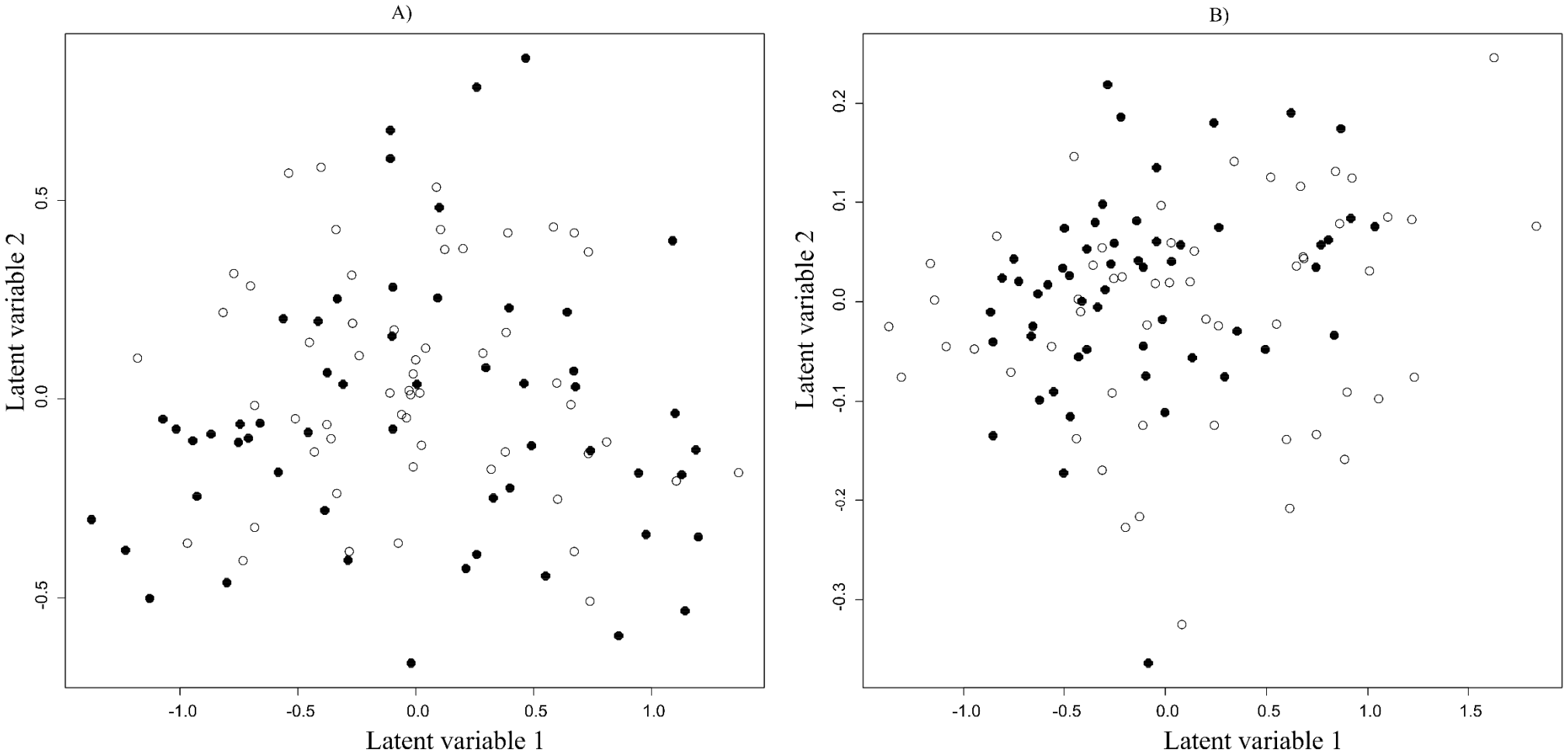


Fig. 4.5 Graphical representation of A) pure latent variable model and B) latent variable model with environmental variables included, showing the similarity of overall dragonfly community between the ENs and PAs. The similarity of assemblages at individual sites is expressed by the distance between the circles within a two-dimensional plane. Open circles = plantation sites, Black circles = natural sites.

4.3.6 Dragonfly species traits

According to the fourth corner trait analyses for ENs only, species with no preference for water body type (habitat generalists) inhabited lower elevations, whereas no elevation preferences were identified for species classified as lentic or lotic occupants. Species that were national endemics, characteristically more sensitive individuals, favoured higher elevations. Species in family Coenagrionidae occurred at sites with fewer rocks but national endemics preferred areas with greater rock cover. Lower water temperatures were favoured by species with no preference for lentic or lotic water bodies and/or were national endemics and/or species from the Synlestidae family (cool-adapter group). Warmer water temperatures were favoured by species that were localised across most of South Africa. Areas where water conductivity was high were preferred by species from the Libellulidae and/or those with no preference for water body type. National endemic species favoured lower water conductivity. The Platycnemididae occurred at highly transformed areas in a 5 km radius. Species that had a preference for dams were found at sites with greater cattle activity (Fig. 4.6 A).

For PA sites, sites with more shade were preferred by shade tolerant species and/or extremely sensitive species. Sites with less shade cover were preferred by low elevation species. Less turbid water was favoured by species that were on the wing for a longer period of the year and/or species from the family Synlestidae and/or lentic species and/or those that did not have a lifestyle preference for perching or flying and/species that were not shade tolerant. Sites with high rock cover were favoured by species with higher elevation ranges. Sites with lower rock cover were favoured by species classified as flyers. Species without a preference for water body type occurred at lower elevations. National endemic species occurred at high elevations and were associated with slopes. Slopes were also preferred by extremely sensitive species and/or those from the family Synlestidae (Fig. 4.6 B).

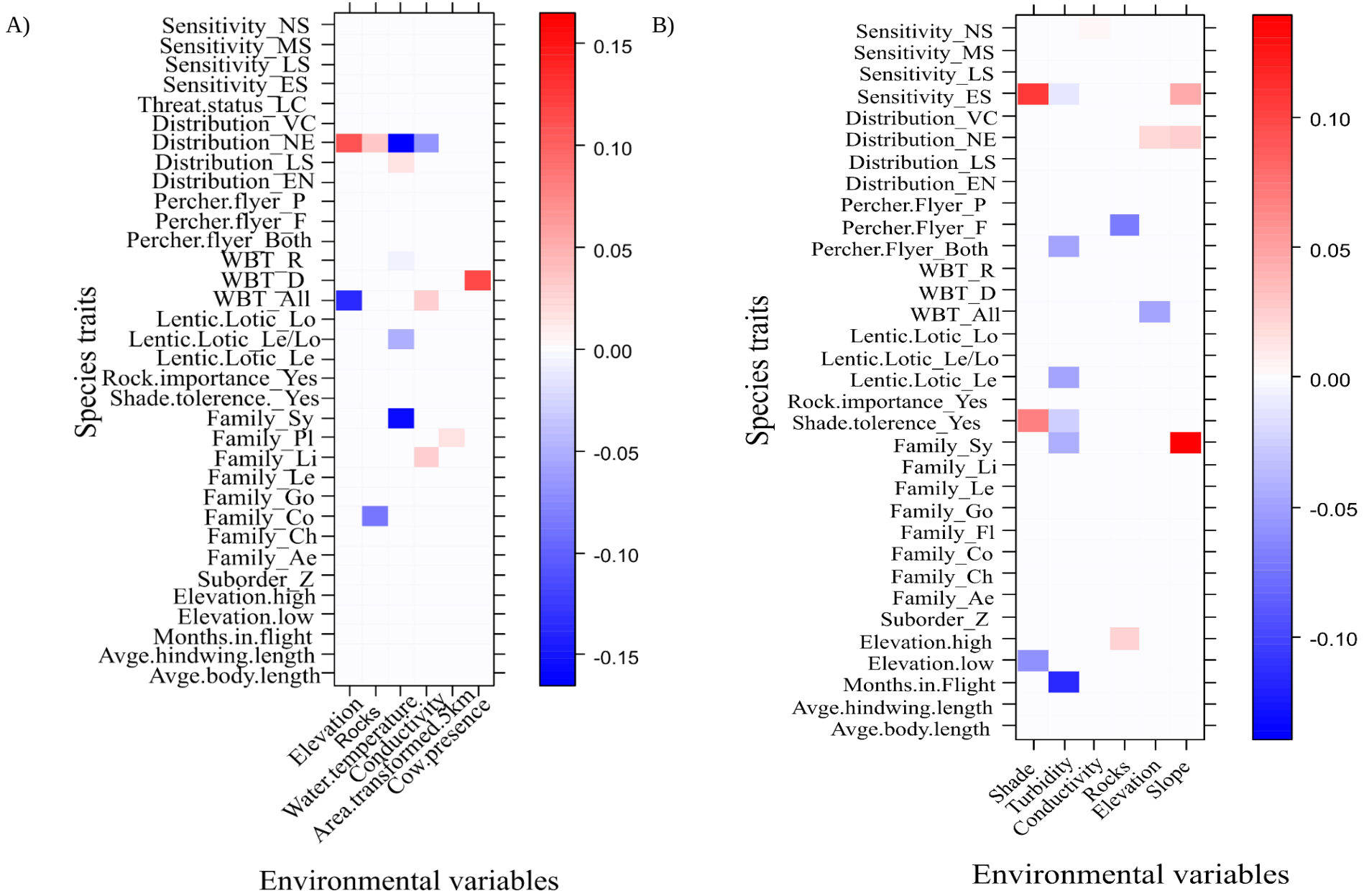


Fig. 4.6 Fourth corner models of species traits showing only the significant environmental variables for A) plantation sites and B) natural sites.

4.4 Discussion

Most importantly, this study showed that there were no significant differences in dragonfly abundance, species richness, DBI scores or assemblage composition when rivers were situated either in ecological network (EN) conservation corridors or were surrounded by large areas of natural grassland of the protected areas (PAs). Rivers within these corridors are therefore of the same quality as natural, undisturbed rivers in terms of the sensitive dragonfly assemblages and general river ecosystem integrity. This is very encouraging for the future of sustainable production landscapes, as these lotic results support those in the same geographical area on the lentic water bodies of wallows, ponds and marshes, where 75% of dragonfly species were shared between ENs and PAs (Pryke et al., 2015). It also supports findings on great similarity in species richness and assemblage composition of various terrestrial arthropods (Formicidae, Araneae, Orthoptera, Lepidoptera and Scarabaeidae) in ENs vs. PAs (Pryke & Samways, 2012), as well as among plants in ENs vs. PAs (Joubert & Samways, 2014). In sum, these ENs composed of large-scale conservation corridors are highly effective in mitigating the effects of fragmentation brought on by plantation forestry, and are being sufficiently well-managed to ensure connectivity between habitat patches for a range of biota.

The width of conservation corridors within ENs was an important environmental variable for dragonfly abundance and richness. Wider corridors characteristically have higher habitat heterogeneity (Haddad et al., 2003). This means that they would fulfil the habitat requirements of a greater number of dragonfly species, supporting a higher species richness. Larger areas of natural habitat also have a greater amount of resources available, which may be why they supported a higher abundance of dragonflies. Many male dragonflies are highly territorial and larger corridors would provide more space for a greater number of potential territories (Harvey & Corbet, 1985). Maximising corridor width should therefore be a priority when designing corridors. However, site DBI scores were not influenced by corridor width. This may be due to majority of the plantation trees in these corridors already adhering to the minimum recommended distance of 30 m away from the rivers' edges. It is possible that these are already wide enough to reduce the influences that would otherwise decrease river quality.

For dragonfly assemblages in plantation and natural sites, management variables related to anthropogenic disturbances had less of an effect on assemblages compared to natural variables. Water variables were the most important drivers of the dragonfly assemblages. This

corresponds with findings elsewhere in South Africa (Western Cape Province), where heterogeneity of in-water variables was the major driver of dragonfly assemblages in untransformed river reaches (Kietzka et al., 2017). However, when the effects of transformation are severe, other variables related to anthropogenic disturbance may override the more natural drivers of heterogeneity in shaping dragonfly assemblages. For example, in the same geographical as here, but before the current environmental management protocols were fully implemented, dragonfly assemblages were strongly influenced by shading from timber trees adjacent to the rivers, which greatly decreased dragonfly diversity (Kinvig & Samways, 2000). This contrasts with the findings here, where no plantation trees were adjacent to the water, clearly indicating that the current management approach to have a substantial riparian corridor of natural vegetation, as recommended by Samways and Steytler (1996) and Kinvig and Samways (2000), is highly effective in maintaining dragonfly assemblages. One of the reasons for this is that the influence of plantation tree shading, a highly significant adverse variable, has been mitigated against. Once the riparian corridor is restored, the most significant variables are those of in-water and elevation, as shown here.

Presence of alien plant species decreased species richness and DBI scores in both PAs and ENs. The effects of alien plants species on dragonflies are highly detrimental, and often related to shading by alien trees (Samways & Taylor, 2004; Samways & Grant, 2006). Here, for dragonfly assemblage composition, shading from alien trees was not the main influence, which may be an indication of their successful removal. However, overall assemblage composition was influenced by the presence of the alien plant species, *C. vulgare*. Although an alien, *C. vulgare* is in the Asteraceae, and would not shade out dragonfly habitats. Its significant influence on the dragonfly assemblage composition here may have been due to chance, although, being a spiky plant, may have had had an inhibitory effect, as the local dragonflies are highly susceptible to entanglement by spiky or hooked plants (Samways, 1991). However, this goes against findings from California, where alien *C. vulgare* positively affected native dragonfly species by the provision of predation refuges (Grof-Tisza et al., 2017).

Presence of cattle was the only other management variable that significantly affected dragonfly assemblages at plantation sites. Impacts from cattle can vary, and depend on their density and behaviour. Some dragonfly species, such as *Chlorolestes fasciatus*, can be present where cattle, as well as some invasive alien plants, are present (Kinvig & Samways, 2000). However, cattle trampling can cause significant damage to the riverbanks, which can threaten some rare local

dragonflies, and is one of the threatening factors for *C. apricans* (Samways, 1995), recorded here. The fourth corner model for plantations showed species that preferred dams and pools occurred where cattle were also present. This may be due to cattle trampling causing siltation at certain river sections and creating still pools, which become inhabited by dragonflies with a preference for that habitat type. Alternatively, cattle may prefer to drink at pools along a river because they are easier to drink from compared to fast flowing water and certain dragonfly species have a preference for those same areas.

At the PA sites, one additional species, the generalist *Pseudagrion hageni*, was recorded that was absent from EN sites, whereas an additional four species were recorded at EN sites yet absent from the PA sites. Interestingly, *C. apricans* was only recorded in plantations, and was the only species in this study with the highest possible DBI score of nine. This is the first record of this species outside of its very narrow previous range in the Amatola-Winterberg, Eastern Cape Province. This species is near endemic (now confined to only two South African provinces), endangered and highly sensitive to habitat change (Samways & Simaika, 2016). Rivers within well-managed corridors can therefore conserve both generalist and highly sensitive, specialist species.

Although DBI site scores did not differ between plantation and natural sites, for both land use types, scores decreases with an increase in the area of transformed land within a 1 km radius. This indicates the potential of the DBI to perform not only at the site scale but also at larger spatial scales. In terms of species traits, some traits could be related to a specific land-use type. Species that are extremely sensitive to disturbance and are national endemics, favoured areas of high elevation and with slopes. Species with no preference for a particular water body type, i.e. habitat generalists, occurred at lower elevations, which were more characteristic of the plantation areas.

4.5 Conclusion

I have shown that the instigation of ENs for conserving biodiversity within plantation forests is an effective mitigation measure that benefits not only terrestrial biodiversity (Pryke & Samways, 2012; van Schalkwyk et al., 2017) but also the semi-aquatic insect order, Odonata. Furthermore, there were no differences in DBI scores between plantation and natural sites, which means the freshwater conditions were similar among sites in the ENs and PAs. Not only

are well-managed ENs conserving dragonfly diversity, but are also ensuring water conditions comparable to those of PAs, which will ideally result in the conservation of other aquatic taxa, as dragonflies are a good umbrella taxon. However, the conservation corridors making up the ENs need to be wide enough to mitigate the negative influences that plantation trees have on river ecosystems, such as shading and water consumption. The importance of maintaining the natural heterogeneity of in-water parameters was highlighted here. Therefore, to maintain river ecosystem integrity, there should be avoidance of disturbances that could alter water chemistry. For example, cattle can have strong effects on water quality, so it is crucial to ensure that stocking rates are kept at low to moderate levels. The removal of any invasive plants should also be a prime focus, as their effects on water quality and quantity can be severe, which was evident from decreased species richness and DBI scores with increased alien plant presence.

4.6 References

Basiron, Y. (2007). Palm oil production through sustainable plantations. European Journal of Lipid Science and Technology, 109(4), 289–295.

Bates, D., Maechler, M., Bolker, B., & Walker, S. (2014). lme4: Linear mixed–effects models using Eigen and S4. R package version, 1(7), 1–23.

Bazelet, C. S., & Samways, M. J. (2011). Relative importance of management vs. design for implementation of large–scale ecological networks. Landscape Ecology, 26(3), 341–353.

Bennett, A. F. (1999). Linkages in the landscape: The role of corridors in wildlife conservation. IUCN, Gland, Switzerland and Cambridge.

Bolker, B. M., Brooks, M. E., Clark, C. J., Geange, S. W., Poulsen, J. R., Stevens, M. H. H., & White, J. S. S. (2009). Generalized linear mixed models: a practical guide for ecology and evolution. Trends in Ecology and Evolution, 24(3), 127–135.

Brown, A. M., Warton, D. I., Andrew, N. R., Binns, M., Cassis, G., & Gibb, H. (2014). The fourth-corner solution–using predictive models to understand how species traits interact with the environment. Methods in Ecology and Evolution, 5(4), 344–352.

Burkey, T. V. (1995). Extinction rates in archipelagos: implications for populations in fragmented habitats. Conservation Biology, 9 (3), 527–541.

Calcagno, V., & de Mazancourt, C. (2010). glmulti: an R package for easy automated model selection with (generalized) linear models. Journal of Statistical Software, 34(12), 1–29.

Cowling, R. M., Pressey, R. L., Rouget, M., & Lombard, A. T. (2003). A conservation plan for a global biodiversity hotspot—the Cape Floristic Region, South Africa. Biological Conservation, 112(1–2), 191–216.

Dudgeon, D., Arthington, A. H., Gessner, M. O., Kawabata, Z.–I., Knowler, D. J., Lévêque, C., & Sullivan, C. A. (2006). Freshwater biodiversity: importance, threats, status and conservation challenges. Biological Reviews, 81(2), 163–182.

Fahrig, L. (1997). Relative effects of habitat loss and fragmentation on population extinction. The Journal of Wildlife Management, 603–610.

Fahrig, L., & Merriam, G. (1994). Conservation of fragmented populations. Conservation Biology, 8, 50–59.

Fedorowick, J. M. (1993). A landscape restoration framework for wildlife and agriculture in the rural landscape. Landscape and Urban Planning, 27(1), 7–17.

Forman, R. T. T. (1995). Some general principles of landscape and regional ecology. Landscape Ecology, 10(3), 133–142.

Forman, R. T. T., & Godron, M. (1986). Landscape Ecology. John Wiley & Sons, New York, USA.

Gittleman, J. L., & Kot, M. (1990). Adaptation: statistics and a null model for estimating phylogenetic effects. Systematic Zoology, 39(3), 227–241.

Grof-Tisza, P., LoPresti, E., Heath, S. K., & Karban, R. (2017). Plant structural complexity and mechanical defenses mediate predator–prey interactions in an odonate–bird system. Ecology and Evolution, 7(5), 1650–1659.

Haddad, N. M. (1999). Corridor and distance effects on interpatch movements: a landscape experiment with butterflies. Ecological Applications, 9(2), 612–622.

Haddad, N. M., Bowne, D. R., Cunningham, A., Danielson, B. J., Levey, D. J., Sargent, S., & Spira, T. (2003). Corridor use by diverse taxa. Ecology, 84(3), 609–615.

Hansen, S., Roets, F., Seymour, C. L., Thébault, E., van Veen, F. J. F., & Pryke, J. S. (2018). Alien plants have greater impact than habitat fragmentation on native insect flower visitation networks. Diversity and Distributions, 24(1), 58–68.

Harabiš, F., & Dolný, A. (2012). Human altered ecosystems: suitable habitats as well as ecological traps for dragonflies (Odonata): the matter of scale. Journal of Insect Conservation, 16(1), 121–130.

Harvey, I. F., & Corbet, P. S. (1985). Territorial behaviour of larvae enhances mating success of male dragonflies. Animal Behaviour, 33(2), 561–565.

Howe, R. W., Davis, G. J., & Mosca, V. (1991). The demographic significance of 'sink' populations. Biological Conservation, 57, 239–255.

Hui, F. K. C. (2016). BORAL – Bayesian ordination and regression analysis of multivariate abundance data in R. Methods in Ecology and Evolution, 7, 744–750.

Jongman, R. H. G. (2002). Homogenisation and fragmentation of the European landscape: ecological consequences and solutions. Landscape and Urban Planning, 58(2–4), 211–221.

Jongman, R. H. G., & Pungetti, G. (2004). Ecological Networks: Concept, Design, Implementation. Cambridge University Press, Cambridge, UK.

Joubert, L., Pryke, J. S., & Samways, M. J. (2017). Moderate grazing sustains plant diversity in Afromontane grassland. Applied Vegetation Science, 20(3), 340–351.

Joubert, L., & Samways, M. J. (2014). Equivalence of grasslands in an ecological network and a World Heritage Site. Biodiversity and Conservation, 23(10), 2415–2426.

Kietzka, G. J., Pryke, J. S., & Samways, M. J. (2015). Landscape ecological networks are successful in supporting a diverse dragonfly assemblage. Insect Conservation and Diversity, 8(3), 229–237.

Kietzka, G. J., Pryke, J. S., & Samways, M. J. (2017). Aerial adult dragonflies are highly sensitive to in–water conditions across an ancient landscape. Diversity and Distributions, 23(1), 14–26.

Kinvig, R. G., & Samways, M. J. (2000). Conserving dragonflies (Odonata) along streams running through commercial forestry. Odonatologica, 29(3), 195–208.

LaRue, M. A., & Nielsen, C. K. (2008). Modelling potential dispersal corridors for cougars in midwestern North America using least–cost path methods. Ecological Modelling, 212(3–4), 372–381.

Lemckert, F., Brassil, T., & Towerton, A. (2005). Native vegetation corridors in exotic pine plantations provide long-term habitat for frogs. Ecological Management and Restoration, 6(2), 132–134.

Lepš, J., & Šmilauer, P. (2003). Multivariate analysis of ecological data using CANOCO. Cambridge University Press, Cambridge, UK.

Lipsey, M. K., & Hockey, P. A. R. (2010). Do ecological networks in South African commercial forests benefit grassland birds? A case study of a pine plantation in KwaZulu–Natal. Agriculture, Ecosystems and Environment, 137(1–2), 133–142.

MacArthur, R. H., & Wilson, E. O. (1967). The theory of Island Biogeography. Princeton University Press, Princeton, UK.

MacDonald, M. A. (2003). The role of corridors in biodiversity conservation in production forest landscapes: a literature review. Tasforests, 14, 41–52.

Mucina, L., & Rutherford, M. C. (2006). The vegetation of South Africa, Lesotho and Swaziland. South African National Biodiversity Institute, Pretoria, South Africa.

Noss, R. F. (1987). Corridors in real landscapes: a reply to Simberloff and Cox. Conservation Biology, 1(2), 159–164.

Noss, R. F., & Harris, L. D. (1986). Nodes, networks, and MUMs: Preserving diversity at all scales. Environmental Management, 10, 299– 309.

Oberg, A. L., & Mahoney, D. W. (2007). Linear mixed effects models. In Topics in biostatistics. Humana Press (pp. 213–234), New York, USA.

Oksanen, J., Kindt, R., Legendre, P., O'Hara, B., Simpson, G.L., Solymos, P., Stevens, M.H. and Wagner, H. (2008). The vegan package version 1.15–0. Available: http://cran. r–project. org/, http://vegan. r–forge. rproject. org/(27.04. 2009).

Pryke, S. R., & Samways, M. J. (2001). Width of grassland linkages for the conservation of butterflies in South African afforested areas. Biological Conservation, 101(1), 85–96.

Pryke, J. S., & Samways, M. J. (2012). Ecological networks act as extensions of protected areas for arthropod biodiversity conservation. Journal of Applied Ecology, 49(3), 591–600.

Pryke, J. S., & Samways, M. J. (2015). Conserving natural heterogeneity is crucial for designing effective ecological networks. Landscape Ecology, 30(4), 595–607.

Pryke, J. S., Samways, M. J., & De Saedeleer, K. (2015). An ecological network is as good as a major protected area for conserving dragonflies. Biological Conservation, 191, 537–545.

R Development Core Team. (2016). R: A language and environment for statistical computing. Vienna: R Foundation for Statistical Computing. Available: http://www.R–project.org. (01/06/2016).

Rees, J. D., Kingsford, R. T., & Letnic, M. (2018). Changes in desert avifauna associated with the functional extinction of a terrestrial top predator. Ecography, 42(1), 67 – 76.

Rodenhouse, N. L., Barrett, G. W., Zimmerman, D. M., & Kemp, J. C. (1992). Effects of uncultivated corridors on arthropod abundances and crop yields in soybean agroecosystems. Agriculture, Ecosystems and Environment, 38(3), 179–191.

Rosenberg, D. K., Noon, B. R. & Meslow, E. C. (1997). Biological corridors: form, function and efficiency. BioScience, 47, 677–687.

Royston, J. P. (1995). Shapiro–Wilk normality test and P–value. Applied statistics, 44(4), 547–551.

Samways, M. J. (1991). Dragonfly death by entanglement on hooked and barbed plant surfaces (Anisoptera: Libellulidae). Odonatologica, 20(1), 79–83.

Samways, M. J. (1995). Conservation of the threatened, endemic dragonflies of South Africa. In proceedings of the international symposium on the conservation of dragonflies and their habitats. Japanese Society for the Preservation of Birds (pp. 8–15), Kushiro, Japan.

Samways, M. J. (2007). Implementing ecological networks for conserving insect and other biodiversity. Insect Conservation Biology, 127–143.

Samways, M. J. (2008). Dragonflies and damselflies of South Africa. Pensoft, Sofia, Bulgaria.

Samways, M. J., Bazelet, C. S., & Pryke, J. S. (2010). Provision of ecosystem services by large scale corridors and ecological networks. Biodiversity and Conservation, 19(10), 2949–2962.

Samways, M. J., & Grant, P. B. C. (2006). Regional response of Odonata to river systems impacted by and cleared of invasive alien trees. Odonatologica, 35(3), 297–303.

Samways, M. J., & Pryke, J. S. (2016). Large–scale ecological networks do work in an ecologically complex biodiversity hotspot. Ambio, 45(2), 161–172.

Samways, M. J., & Simaika, J. P. (2016). Manual of freshwater assessment for South Africa: Dragonfly biotic index. South African National Biodiversity Institute, Pretoria, South Africa.

Samways, M. J., & Steytler, N. S. (1996). Dragonfly (Odonata) distribution patterns in urban and forest landscapes, and recommendations for riparian management. Biological Conservation, 78(3), 279–288.

Samways, M. J., & Taylor, S. (2004). Impacts of invasive alien plants on Red–Listed South African dragonflies (Odonata). South African Journal of Science, 100, 78–80.

Simaika, J. P. & Samways, M. J. (2009). An easy–to–use index of ecological integrity for prioritizing freshwater sites and for assessing habitat quality. Biodiversity and Conservation, 18, 1171–1185.

Simaika, J. P., & Samways, M. J. (2011). Comparative assessment of indices of freshwater habitat conditions using different invertebrate taxon sets. Ecological Indicators, 11(2), 370–378.

Simaika, J. P., & Samways, M. J. (2012). Using dragonflies to monitor and prioritize lotic systems: a South African perspective. Organisms Diversity and Evolution, 12(3), 251–259.

Simberloff, D., & Abele, L. (1976). Island biogeography theory and conservation practice. Science, 191(4224), 285–286.

Simberloff, D., & Cox, J. (1987). Consequences and costs of conservation corridors. Conservation Biology, 1(1), 63–71.

Solt, M. F., Solt, F., Hu, Y., Keyes, O., Bolker, B., Müller, S., & Leeper, T. (2018). Package dotwhisker– Title Dot–and–Whisker Plots of Regression Results.

Stein, J. L., Stein, J. A., & Nix, H. A. (2002). Spatial analysis of anthropogenic river disturbance at regional and continental scales: identifying the wild rivers of Australia. Landscape and Urban Planning, 60(1), 1–25.

ter Braak, C. J. (1990). Update notes: CANOCO, version 3.10. Agricultural Mathematics Group, Wageningen, Holland.

ter Braak, C. J., & Šmilauer, P. (2012). Canoco reference manual and user's guide: software for ordination, version 5.0. Microcomputer power, Ithaca, New York.

van Schalkwyk, J., Pryke, J. S., & Samways, M. J. (2017). Wide corridors with much environmental heterogeneity best conserve high dung beetle and ant diversity. Biodiversity and Conservation, 26(5), 1243–1256.

Vitousek, P. M., Mooney, H. A., Lubchenco, J., & Melillo, J. M. (1997). Human domination of Earth's ecosystems. Science, 277(5325), 494–499.

Wang, Y. I., Naumann, U., Wright, S. T., & Warton, D. I. (2012). mvabund–an R package for model-based analysis of multivariate abundance data. Methods in Ecology and Evolution, 3(3), 471–474.

Wang, Y., Naumann, U., Wright, S., Eddelbuettel, D., & Warton, D. (2014). mvabund: Statistical methods for analysing multivariate abundance data. R package version, 3(3).

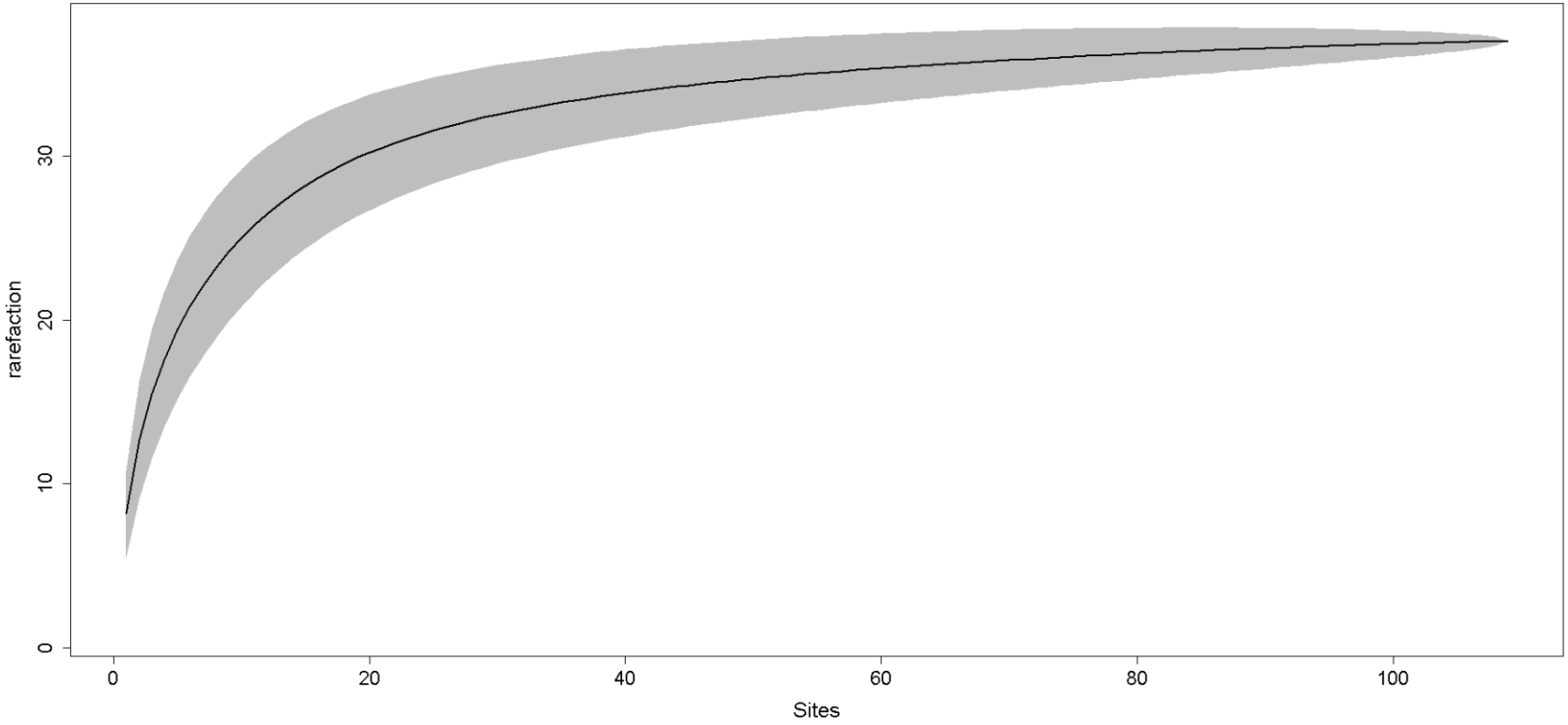
Warton, D.I., Shipley, B., & Hastie, T. (2015). CATS regression – a model–based approach to studying trait–based community assembly. Methods in Ecology and Evolution, 6, 389–398.

Wilcove, D. S., McLellan, C. H., & Dobson, A. P. (1986). Habitat fragmentation in the temperate zone. Conservation Biology, 6, 237–256.

4.7 Appendices

Appendix 4.A Spec ies list and DBI scores of recorded dragonfly species.

Scientific name	DBI
Suborder ZYGOPTERA	
Family CHLOROCYPHIDAE	
Platycypha calligata (Selys 1853)	2
Platycypha fitzsimonsi (Pinhey 1950)	4
Family SYNLESTIDAE	
Chlorolestes apricans (Wilmot 1975)	9
Chlorolestes fasciatus (Burmeister 1839)	4
Family LESTIDAE	
Lestes plagiatus (Burmeister 1839)	2
Family PLATYCNEMIDIDAE	
Allocnemis leucosticta (Selys 1863)	5
Family PROTONURIDAE	
Elattoneura glauca (Selys 1862)	1
Family COENAGRIONIDAE	
Ceriagrion glabrum (Burmeister 1839)	0
Pseudagrion caffrum (Burmeister 1839)	5
Pseudagrion citricola (Barnard 1937)	3
Pseudagion hageni tropicanum(Pinhey 1966)	2
Pseudagrion kersteni (Gersaecker 1869)	1
Pseudagrion salisburyense (Ris 1921)	1
Pseudagrion spernatum (Ris 1921)	3
Ischnura senegalensis (Rambur 1842)	0
Africallagma glaucum (Burmeister 1839)	1
Africallagma sapphirinum (Pinhey 1950)	4
Suborder ANISOPTERA	
Family AESHNIDAE	
Pinheyaeschna subpupillata (McLachlan 1896)	4
Zosteraeschna minuscula (McLachlan 1896)	5
Anax imperator (Rambur 1842)	1
Anax speratus (Hagen 1867)	2
Family GOMPHIDAE	
Notogomphus praetorius (Selys 1878)	4
Ceratogomphus pictus (Selys 1854)	2
Paragomphus cognatus (Rambur 1842)	1
Family LIBELLULIDAE	
Orthetrum caffrum (Burmeister 1839)	3
Orthetrum Julia falsum (Longfield 1955)	1
Nesciothemis farinosa (Förster 1898)	1
Crocothemis erythraea (Brullé 1832)	0
Crocothemis sanguinolenta (Burmeister 1839)	3
Sympetrum fonscolombii (Selys 1841)	0
Trithemis arteriosa (Burmeister 1839)	0
Trithemis dorsalis (Rambur 1842)	0
Trithemis furva (Karsch 1899)	0
Trithemis stictica (Burmeister 1839)	1
Zygonyx natalensis (Martin 1900)	2
Pantala flavescens (Fabricius 1798)	0
Tramea basilaris I(Palisot de Beauvois 1817)	0



Appendix 4.B Rarefied species accumulation curve for overall dragonflies.

Appendix 4.C Summary statistics for environmental variables between plantation and natural sites.

Plantation	Min.	1st Qu.	Median	Mean	3rd Qu.	Max.	Natural	Min.	1st Qu.	Median	Mean	3rd Qu.	Max.
Cow presence	0	0	0	20.45	40	100	Cow presence	0	0	0	11.09	0	100
Shade (%)	0	0	30	35.64	60	90	Shade (%)	0	0	5	21.82	40	90
Pool (%)	0	0	0	19.27	35	100	Pool (%)	0	0	0	13.64	15	90
Riffle (%)	0	0	20	26.73	50	100	Riffle (%)	0	0	30	38.36	70	100
Glide (%)	0	20	50	51.27	85	100	Glide (%)	0	10	50	47.82	85	100
Width (m)	0.3	1	2	3.285	3.5	20	Width (m)	0.3	1	4	9.002	10	80
Depth (m)	0.1	0.3	0.4	0.48	0.5	3	Depth (m)	0.1	0.25	0.6	1.137	1.75	5
Turbidity (NTU)	1	1	2	2.236	3	5	Turbidity (NTU)	1	1	2	2.473	3.5	5
Temp (°C)	16.1	19.3	21.7	21.5	22.7	28.6	Temp (°C)	16.7	20.05	21.9	23.27	24.05	28.4
pH	6.15	7.605	7.75	7.708	7.89	8.69	pH	6.98	7.54	7.77	7.801	8.065	8.83
Cond (ppt)	0.02	0.03	0.03	0.04018	0.045	0.14	Cond (ppt)	0.01	0.02	0.03	0.03382	0.04	0.16
Dis Ox (%)	0.07	72.15	76.2	75.83	80.9	112	Dis Ox (%)	42.8	73.4	79.2	77.13	82.75	92.3
Sand (%)	0	0	0	12.91	10	100	Sand (%)	0	0	10	25.45	50	100
Rock (%)	0	0	0	11.82	15	90	Rock (%)	0	0	0	7.091	5	80
Debris (%)	0	0	0	2.545	0	40	Debris (%)	0	0	0	0.6364	0	10
R.cuneifolius (%)	0	0	10	22.18	40	100	*R.cuneifolius* (%)	0	0	0	7.636	2.5	80
S. mauritianum (%)	0	0	0	1.364	0	20	*S. mauritianum* (%)	0	0	0	0.3636	0	10
C. vulgare (%)	0	0	0	3.636	0	40	*C. vulgare* (%)	0	0	0	0.8182	0	30
Other invasives	0	0	0	17.18	30	80	Other invasives	0	0	0	10.18	0	100
Veg height (%)	0.2	1	1.2	1.956	2.25	8	Veg height (%)	0	0.9	1.5	3.258	3	30
Macrophytes (%)	0	0	0	5.636	0	50	Macrophytes (%)	0	0	0	6.727	0	80
Elevation (m a.s.l)	849	1102	1290	1245	1372	1563	Elevation (m a.s.l)	616	1147	1485	1405	1680	1973
Slope	1	4	6	6.909	9	21	Slope	0	3	6	7.2	10.5	25
Aspect	1	115.5	179	177.6	247	352	Aspect	2	49.5	148	152.2	236.5	353
Corridor width (m)	9	72.5	136	203.9	264	740	Transformed 1km	0	0	0	19451	0	606996
Transformed 1km	113421	627161	1753156	1468031	2155309	3222507	Transformed 5km	0	0	0	113.3	0	1692
Transformed 5km	774955	3187092	9694760	11163743	19237221	25249731							

Appendix 4.D Table showing the effect of environmental variables on species richness and DBI scores. Variables excluded from models shown by /.

Variables		Species richness			DBI scores		
		Overall	Plantation	Natural	Overall	Plantation	Natural
Natural variables	Elevation (m a.s.l)	7.64**	/	4.66*	/	/	/
	Aspect (degrees)	/	4.58*	/	/	/	/
	Slope (%)	4.55*	/	2.51	/	/	/
	Shade (%)	13.61***	/	5.80*	/	/	/
	Sand substratum (%)	/	/	/	/	/	/
	Rock side (%)	/	/	/	/	/	/
	River width (m)	/	/	/	/	/	/
	River depth (m)	/	7.82**	/	/	/	/
	Riffles	3.93*	5.12*	/	/	/	/
In-water measures	Water temperature (˚C)	/	/	/	8.83**	/	18.95***
	Conductivity (ppt)	/	/	/	/	6.18*	/
	pH	/	/	/	/	/	/
	Dissolved oxygen (%)	7.52**	21.84***	/	/	/	/
	Turbidity (NTU)	5.52*	/	/	7.10**	/	/
Anthropogenic variables	EN vs PA	0.44	/	/	/	/	/
	Area transformed (1 km)	/	/	/	/	/	/
	Area transformed (5 km)	6.46*	2.30*	/	/	/	/
	Rubus cuneifolius (%)	/	/	/	/	/	/
	Solanum mauritianum (%)	6.64**	18.17***	/	/	/	/
	Invasive plants (%)	10.16**	13.54***	/	/	/	/
	Corridor width (m)	/	/	/	/	/	/
	Cattle presence (%)	/	/	/	/	/	/

* P < 0.05, ** P < 0.01, *** P < 0.001

Chapter 5: Lessons learnt from thirty years of monitoring a pond designed for dragonfly conservation

Abstract

Pond construction in urban areas can offset loss of aquatic insects by providing refuges. Urban ponds are also a point of interface between the public and aquatic insects, especially the highly charismatic dragonflies. Ponds have therefore been constructed specifically for dragonfly conservation in many countries, yet they require regular management. Often ponds can be constructed along a already existing stream. A major issue for such ponds is vegetation overgrowth and natural infilling from both organic and inorganic material, eventually reverting the pond to a stream, which is unsuitable for many species. Here, changes in dragonfly diversity were assessed over a 30-year period at a pond constructed for aquatic insect conservation in the Pietermaritzburg Botanical Gardens, South Africa. Adult, male dragonflies and 13 environmental variables were recorded along 31 transects during summer months from 2015 to 2018. This was compared to previous data collected at the same sites before pond construction and in the short- and medium-terms after that. During the years when the pond was present, dragonfly abundance and species richness was higher compared to when it was a stream. After the pond was restored in 2018 it resembled the original pond with a large area of open water, a variety of flow regimes and a high diversity of indigenous plant species. Dragonfly abundance and species richness both increased after the pond was restored, which was strongly linked to decreased vegetation cover and removal of alien plant species. Alien vegetation was also the most important driver of changes in dragonfly assemblages, followed by vegetation cover and grass height. Here, shortly after restoration measures, the dragonfly assemblage had almost completely recovered to that of the original pond. To achieve such results it is essential to produce a variety of microhabitats using selective management of marginal vegetation and to create a range of flow regimes. Thereafter, constant future management is recommended to prevent vegetation overgrowth and siltation of the pond.

5.1 Introduction

Freshwater is a vital resource (Johnson, et. al., 2001) and owing to our dependence on freshwater ecosystems, they are often surrounded by urban landscapes. Due to the anthropogenic pressures placed on these ecosystems, protected areas are crucial for the conservation of aquatic biodiversity (Hill et al., 2016). However, additional refuges within urban landscapes can be provided for aquatic biota through the construction of ponds (Hassall & Anderson, 2015).

Generally, more heterogeneous habitats host greater biodiversity (Frainer et al., 2018), and the construction of a specially-designed pond along a stream in an urban area provides habitats for both lentic and lotic aquatic insects, and so increases overall biodiversity in the local area. Such a pond is, in effect, like a large deposition pool in a stream, and inevitably collects inorganic and organic matter. In 1988, a designer pond was constructed along a stream in the National Botanical Gardens in Pietermaritzburg, KwaZulu-Natal (Kingfisher Lake). The main reason for this pond was to assist with insect and aquatic plant conservation, but importantly, it was designed as a key part of a dragonfly conservation awareness trail (Suh & Samways, 2001). Dragonfly richness and abundance significantly increased as a result of the construction of this pond, through local introduction of a specifically-designed complex lentic system into a lotic one, so increasing habitat diversity substantially (Suh & Samways, 2005). These designer ponds can be planned according to the requirements of specific taxa, as in the case of artificial ponds for dragonflies in Japan (Primack et al., 2000) and Europe (Harabiš & Dolný, 2012; Maynou et al., 2017).

Dragonflies are often considered model organisms for freshwater research (Córdoba-Aguilar 2008; Kietzka et al., 2017). They play important ecological roles as top predatory insects and have life cycles with both an aquatic and a terrestrial phase, with the aerial adults readily dispersing to locate their specific habitats (Corbet, 1999). Dragonflies are widely used as bioindicators, as they are sensitive to many changes in their environments (Briers & Biggs, 2003; Harabiš & Dolný, 2012; Kietzka et al., 2018). The adults are well-researched, and the highly territorial males are easy to sample and identify to species level (Moore, 1991). In South Africa, their success as indicators led to the development of the Dragonfly Biotic Index (DBI), a scoring system which uses the presence of dragonfly at a site to define the health of that freshwater ecosystem (Simaika & Samways, 2009a; b; 2011). More recently, dragonflies have

been recognised as umbrella species, whose biodiversity is reflective of the biodiversity of co-occurring taxa (Bried et al., 2007; Oertli, 2008; Balzan, 2012; Maynou et al., 2017; Chapter 2).

When a pond is constructed within a slow-flowing stream system, there is a change in hydrodynamics. Siltation can lead to reversion back to a stream through in-filling while maintaining a passage for water flow. This structural change brings about successional changes in biota over time (Wood & Armitage, 1997; Salemi et al., 2016; Auerswald & Geist, 2018). Due to their high mobility as adults and sensitivity to their surroundings, dragonfly assemblages associated with a pond are also likely to change over time, following ecological succession patterns. The short- and long-term assemblage changes around constructed conservation ponds have been documented in some European countries (Schmidt 1985; Moore, 1991; 2001; Chovanec, 1994; Chovanec & Raab, 1997).

Suh & Samways (2005) provided recommendations for pond ecosystem management based on the habitat requirements of dragonfly species associated with specific stages of succession. These included the removal of alien plant species, rotational cutting of marginal vegetation, passive establishment of indigenous marginal plant diversity without creating too much shade, and, importantly, occasional dredging of the pond. In response to these recommendations, the pond was heavily dredged and restored in 2017, 29 years after initial construction. It was once again transformed into a large open-water pond with numerous biotopes in terms of vegetation and flow regimes and, after much effort, resembled the one originally designed. However, it was not known whether the restoration of this pond also led to the return of the full complement of dragonfly species.

The main aim here is to assess dragonfly assemblage response to changes in biotopes as a result of resetting of succession through mostly dredging. Specifically, I focus on dragonfly assemblage changes: 1) between the years when the pond was present (both the original pond and the restored pond) compared to when the pond was absent (a stream, before and in-between pond creation and restoration), 2) the reversion of the pond back to stream between 2015 and 2017 (due to both vegetation succession and siltation of the pond), and 3) pond restoration efforts after intensive maintenance in 2018. In this study, I therefore attempt to determine the response of dragonflies to short- and long-term changes in habitat heterogeneity due to biotope creation, biological succession due to lack of proper pond maintenance, and shortly after

maintenance in 2018. The most important variables for maintaining high dragonfly diversity at this artificial conservation pond in an urban setting will therefore be highlighted.

5.2 Materials and methods

5.2.1 Study site and dragonfly sampling

This study was conducted at the National Botanical Gardens in Pietermaritzburg, KwaZulu-Natal, South Africa (-29.604697 S, 30.348078 E, 790 m a.s.l). Two major features of this botanical garden is that it is rich in dragonfly species (Willis & Samways, 2011) and supports a specially designed dragonfly viewing trail for citizen scientists and the public at large (Willis & Samways, 2013). Part of this trail in includes a large man-made pond (Kingfisher Lake).

Assessment of dragonfly species was conducted along the historic stream before the pond was constructed in 1988, and 12 species were recorded. Shortly after the pond was constructed in 1991, dragonfly species richness increased to 26 (Steytler & Samways, 1995). This was followed by gathered information on the short- and medium-term changes at Kingfisher Lake (Steytler & Samways, 1995; Samways et al., 1996; Osborn & Samways, 1996; Suh & Samways, 2001; 2005). The initial, large increase was in response to the creation of the pond within a lotic system, in an area with high dragonfly diversity providing source populations. Additionally, the site is at the edge of an escarpment, and therefore recruits dragonflies from both lower and higher elevations (Suh & Samways, 2005). In 2001, this increased to 30 species due to maintenance of the pond and well-managed vegetation succession (Suh & Samways, 2005). For a poorly managed pond, succession will lead to vegetation overgrowth, establishment of alien plant establishment and pond siltation, as happened in the case of Kingfisher Lake, which reverted back to a narrow, overgrown stream by 2014. This siltation and shading reduced habitat heterogeneity considerably.

Sample units (SUs) in this study were at the same positions as previously used by Steytler and Samways (1995) and Suh and Samways (2005) (Appendix 5.A). Initially, the site was a small stream within an urban landscape in the grassland biome. In June 1989, the stream was impounded and an artificial pond was constructed (550 m circumference) (Suh & Samways, 2005) to create an optimal environment for the conservation of insects and water plants (Steytler & Samways, 1995). It was therefore designed to contain various physical parameters

such as a flowing inlet and outlet, a 3-m high waterfall, and a diversity of other flow regimes (Steytler & Samways, 1995). In an initial survey, a total of 31 SUs were selected, SUs 1 to 27 around the circumference of the pond; 28 to 29 below the outlet; and 30 to 31 were at the stream inlet. Each SU covered an area of 20 m x 2 m, which included 1 m either side of the water's edge (Suh & Samways, 2005). By the year 2014, the pond was completely overgrown and silted up, and only a very small stream was flowing through the area. In 2017, the pond area was dredged, reopened, and alien vegetation was removed. The alien vegetation was replaced with an assortment of indigenous plant species of varying heights to ensure structural and compositional diversity. The project also restored flow regimes that were similar to those present before, and also included three weirs to reduce flow velocity and slow siltation (The Witness, 2018).

Data were extracted from Steytler and Samways (1995) for the years 1988 and 1991, and Suh and Samways (2005) for the years 1993 and 2001. The present study collected data for 2015, 2016, 2017 and 2018. Each SU was sampled six times a year during summer (2015 to 2018) according to the methods used by Suh and Samways (2005). Visual scanning (with the help of binoculars) was used to record the species and abundance of male dragonflies present for six minutes per SU. Observations took place on windless, sunny days, but the mid-day period was avoided as dragonflies are less active when temperatures are too hot (Corbet, 1999). When identification was a challenge, individuals were caught with a net and identified according to Samways and Simaika (2016).

5.2.2 Environmental variables at the sampling unit level

Thirteen environmental variables (EVs) were recorded at each SU from 2015 to 2018, and comprised similar variables as those measured by Suh and Samways (2005). These included: the year of sampling (2015, 2016, 2017 or 2018), flow type (FLOW: pond, stream or no water), the percentage of shade (SH), percentage of water with no emergent vegetation (OW: open water) and percentage cover of vegetation canopy overhanging the water (VEC). The presence or absence of specific vegetation types was also recorded for each SU. Vegetation was categorised into eight groups based on compositional and structural characteristics, as well as on the dominant species. These physiognomic categories were: 1) marginal forest (LUSH): where large tree species dominated, 2) submerged vegetation (SUVG): mostly *Chara* spp., 3) emergent vegetation (EFVG): dominated by waterlilies and *Commelina africana*, 4) herbs, sedges and broad-bladed grass (BUSH): *Cyperus papyrus*, *Ludwigia octovalvis*, *Setaria* spp.,

5) alien plants (EXOT): *Pontederia cordata* and *P. purpureum*, and 6-8) grass height and species: short (SGRA: 0–25 cm) *Paspalum urvillei*, medium (MGRA: 25–50 cm) *Sporobolus pyramidalis* and tall (TGRA: >50 cm).

5.2.3 Statistical analyses

5.2.3.1 Overall influence of pond vs stream on dragonfly species richness

An analysis was conducted for the overall waterbody (i.e. SUs altogether) for the years 1988, 1991, 1993, 2001, 2015, 2016, 2017 and 2018 (n = 1 per year), as individual SU data were not available prior to 1993. Species richness data were extracted from Steytler and Samways (1995) for the years 1988 and 1991, and Suh and Samways (2005) for the years 1993 and 2001. Abundance data were not available from Steytler and Samways (1995). Richness data from 2015 to 2018 were collected in the present study. I divided richness data per collection event (year) into two categories, the years when the system was a pond (1991, 1993, 2001 and 2018), and the years when it was a stream (1988, 2015, 2016 and 2017). Data was normally distributed, therefore a linear model (LM) with Gaussian distribution was run in the *lme4* (Bates et al., 2014) package in R 3.5.0 (R Core Team, 2016) to determine whether dragonfly species richness differed between the pond and stream system states.

5.2.3.2 Changes in dragonfly abundance, species richness and environmental variables over time

Analyses at the SU level (n = 31 per year) were conducted for the years 1993, 2001, 2015, 2016, 2017 and 2018, as there were no data available per SU for species richness and abundance for the years 1988 and 1991. Both dragonfly abundance and species richness data showed non-normal distribution and heterogeneity of variances, even after transformation (Legendre & Legendre, 1998; Pryke & Samways, 2008). Generalized linear mixed-effect models (GLMMs), with SU as the random effect, were run in the *lme4* package (Bates et al., 2014) in R 3.5.0 (R Core Team, 2016) to determine whether dragonfly abundance and species richness were influenced by year of sampling (i.e. between the years when the waterbody was the original pond, a stream or a restored pond). Analyses showed no over-dispersion of variances and were best fitted with a negative binomial distribution with a Laplace approximation (Bolker et al., 2009). Thereafter, Tukey post-hoc tests in the *R* package *multcomp* were used to determine the pairwise differences between the sampling years (Hothorn et al., 2008).

To determine whether the EVs differed between the sampling years, GLMMs and Tukey post-hoc tests were run for the variables recorded from 2015 to 2018. Presence/absence data were fitted with binomial distribution and the remaining variables best fitted Poisson distribution.

5.2.3.3 Effect of environmental variables on dragonfly abundance and species richness

In R (R Core Team, 2016), the *vegan* package (Oksanen et al., 2008) was used to produce a rarefaction curve to determine whether sampling effort was sufficient to capture a true representation of all of the species present.

Analyses of the recorded EVs at the SU level only included the years 2015 to 2018. Correlations between the EVs were calculated using the Spearman's rank order coefficient in R (R core team, 2016) using the *boot* package (Canty & Ripley, 2012). In the event that any two variables were strongly correlated ($r > 0.5$), one was carefully chosen and the other removed.

The SUs were spatially auto-correlated according to Moran's I autocorrelation index, and thus SU was used as a random variable for all analyses (Gittleman & Kot, 1990). As dragonfly abundance and species richness data were non-normal, GLMMs were used to identify influences of the environmental variables on the data. The best fit models were determined using the stepwise forward selection method based on AIC values using the *AICcmodavg* package (Mazerolle & Mazerolle, 2017). Data were fitted with negative binomial distribution for dragonfly abundance data and Poisson error distribution for dragonfly species richness (Bolker et al., 2009). These were performed using the *lme4* package (Bates et al., 2014) in R (R Core Team, 2016). Analyses showed no over-dispersion of variances and thus χ^2- and P-values were calculated (Bolker et al., 2009). For dragonfly species richness, which was significantly influenced by vegetation cover, a piecewise regression test was performed to determine the breakpoints in the *segmented* package in R (Toms & Lesperance, 2003).

5.2.3.4 Effect of environmental variables on dragonfly assemblage composition

A Canonical Correspondence Analysis (CCA) was used to correlate dragonfly assemblage composition data with the recorded environmental variables in CANOCO 5 (ter Braak & Šmilauer, 2012). This was done at the SU level for the years 2015 to 2018. In order to avoid a type 3 error, which is the likelihood of correlated EVs giving a false positive, interactive

forward selection analysis (ter Braak, 1990) was used to select the group of variables that best explained the variation in dragonfly data. This method accounts for covariant and incomplete environmental variables, as well as skewed species distributions (Palmer, 1993). Selection was stopped when there was no significant increase in explained assemblage variation, tested by Monte Carlo permutation. The analysis was permutated 499 times to normalize distribution and allow comparisons of variables (Lepš & Šmilauer, 2003). This was compared to the results from a CCA conducted for the original pond in 2001 (Suh & Samways, 2005).

5.3 Results

5.3.1 Overall influence of pond vs stream on dragonfly species richness

Significantly more species occurred during the times when the pond was present compared to when it was a stream ($F = 31.70$; $p < 0.01$) (Fig. 5.1).

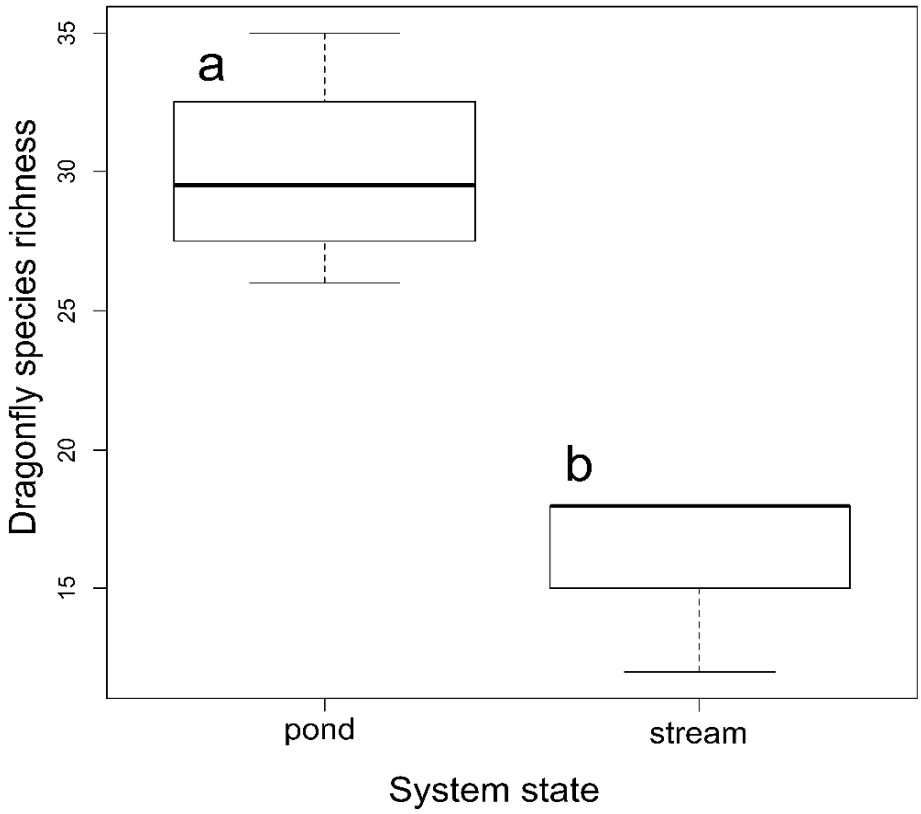


Fig. 5.1 Dragonfly species richness between the years when the system was an open pond and when it was a closed stream.

5.3.2 Changes in dragonfly abundance, species richness and environmental variables over time

Dragonfly abundance ($\chi^2 = 40.62$; $P < 0.001$) and species richness ($\chi^2 = 136.08$; $P < 0.001$) changed significantly over the years due to pond creation, reversion to a stream and pond restoration (Fig. 5.2). There were no significant differences for dragonfly abundance between the years 1993, 2001 and 2018. This is when dragonfly abundance was at its highest, which coincided with the years when the pond was first created (1993 and 2001) and in 2018 after it was restored (Fig. 5.2). The highest dragonfly species richness occurred in 1993 and 2001 also when the pond was first created. This was followed by species richness of the restored pond in 2018. From 2015 to 2017, dragonfly abundance and species richness were at their lowest, when the system was a stream.

The percentage of vegetation cover and open water, did not differ between 2015, 2016 and 2017 when the waterbody was a stream. However, in 2018, after the pond was restored, there was significantly less vegetation cover and more open water than any three of the previous years (Table 5.1; Appendix 5.B). The years 2015 and 2018 had significantly less shade than 2016 and 2017. In 2018, more 'still water' was present compared to the other years. During 2015 and 2016, there were significantly more alien plant species than in 2017 and 2018. However, in 2018, significantly fewer alien species were present than were recorded for the three other years. The presence of marginal indigenous forest, emergent and submergent vegetation, and sedges and herbs, did not vary between the years. Grass of all heights also significantly differed between the years.

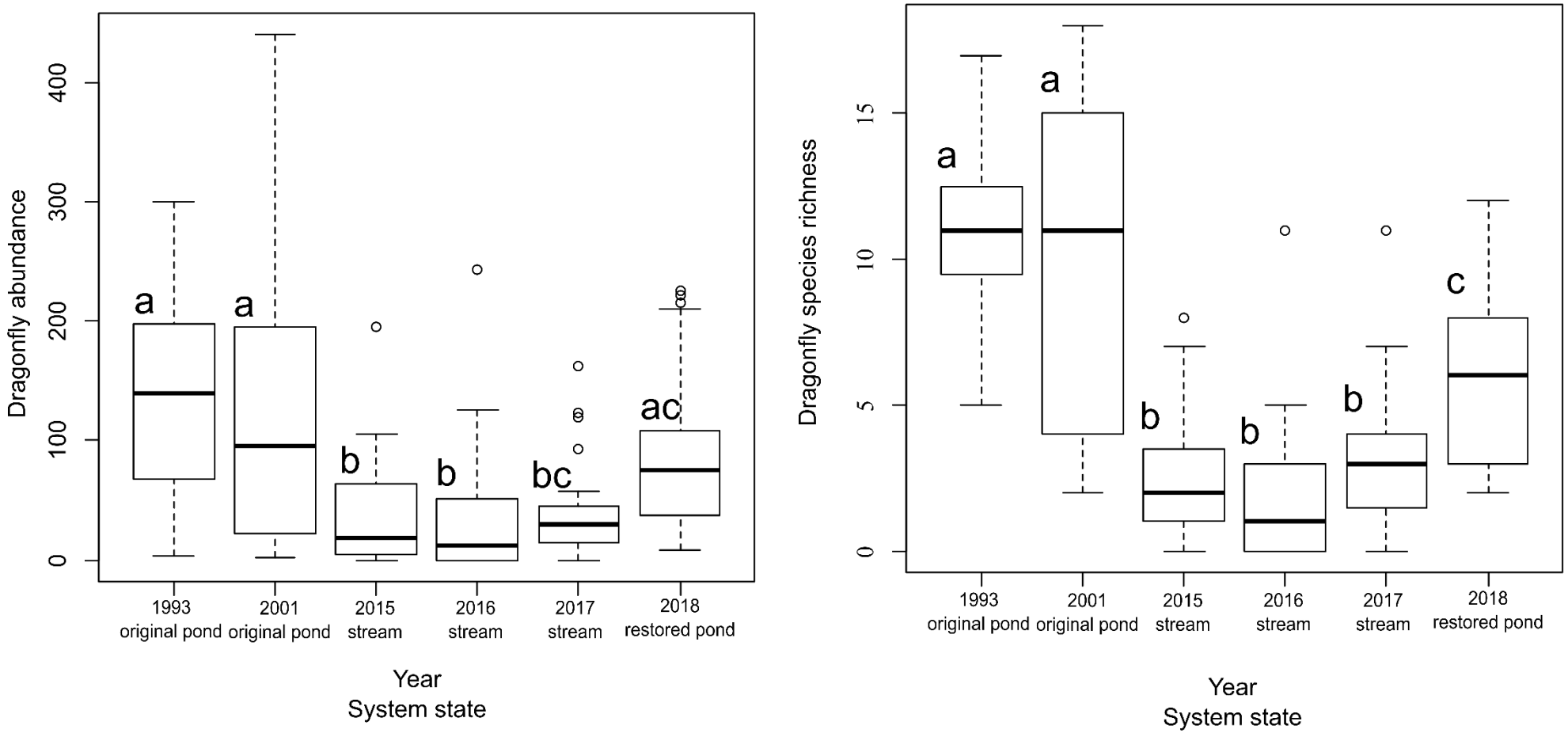


Fig. 5.2 Dragonfly abundance and species richness between the years when the waterbody was the original pond, stream and restored pond.

Table 5.1 The effect of sampling year on environmental variables. Test-statistics are displayed as $\chi 2$ values.

Variables	**$\chi 2$ values**
	Year
Vegetation cover (%)	18.28***
Open water (%)	14.91***
Shade (%)	38.62***
Flow type (P/A)	64.35***
Marginal forest (P/A)	0.38
Emergent vegetation (P/A)	1.74
Submergent vegetation (P/A)	3.61
Herbs, sedges and broad-bladed grasses (P/A)	7.51
Alien vegetation (P/A)	43.64***
Short grass (P/A)	32.62***
Medium grass (P/A)	28.00***
Tall grass (P/A)	30.55***

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

5.3.3 Effect of environmental variables on dragonfly abundance and species richness

The species rarefaction curve reached asymptote, signifying that sampling effort was sufficient and reflective of all the dragonfly species present (Appendix 5.C). Alien plant species and vegetation cover had the greatest influences on dragonfly abundance and species richness (Table 5.2). An increase in vegetation cover decreased dragonfly abundance (Fig. 5.3). The piecewise regression showed that species richness showed a positive correlation to vegetation cover up to a certain point (38.96% +/- 2.75). Vegetation cover was negatively correlated with percentage of open water. This means that as vegetation cover decreased, the amount of open water increased, and so did dragonfly abundance and species richness. Vegetation cover was positively correlated to flow regime and submergent vegetation presence. As vegetation cover increased, there were fewer open pond areas and more streams and areas without any water, which caused significant decreases in dragonfly abundance and species richness. Similarly, an increase in vegetation cover was positively correlated with the presence of submergent vegetation, which significantly decreased dragonfly abundance and species richness. The presence of alien plant species significantly decreased dragonfly abundance and species richness (Fig. 5.4). Alien vegetation presence was also positively correlated with the presence of tall grass (>50 cm), while negatively correlated to the presence of short grass. An increase in shade cover significantly decreased dragonfly species richness (Fig. 5.3).

Table 5.2 Influence of environmental variables on dragonfly abundance and species richness.Test statistics are displayed as $\chi 2$ values.

Variables	**$\chi 2$ values**	
	Abundance	**Species richness**
Vegetation cover (%)	14.07***	32.91***
Alien vegetation (P/A)	4338.00***	22.20***
Shade (%)		5.43*
Herbs, sedges and broad-bladed grasses (P/A)		3.78

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

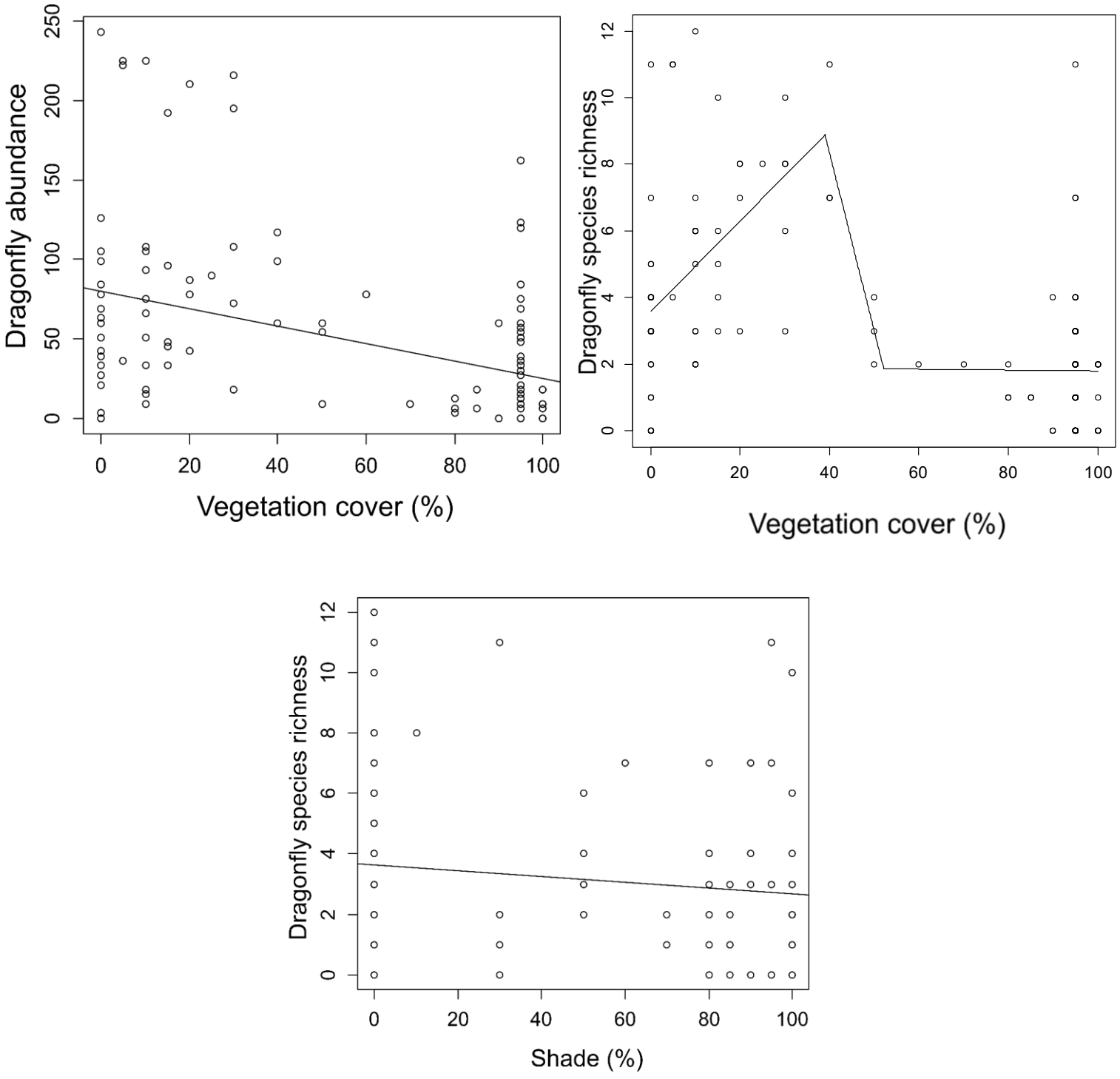


Fig. 5.3 Effect of vegetation cover on dragonfly abundance and species richness.

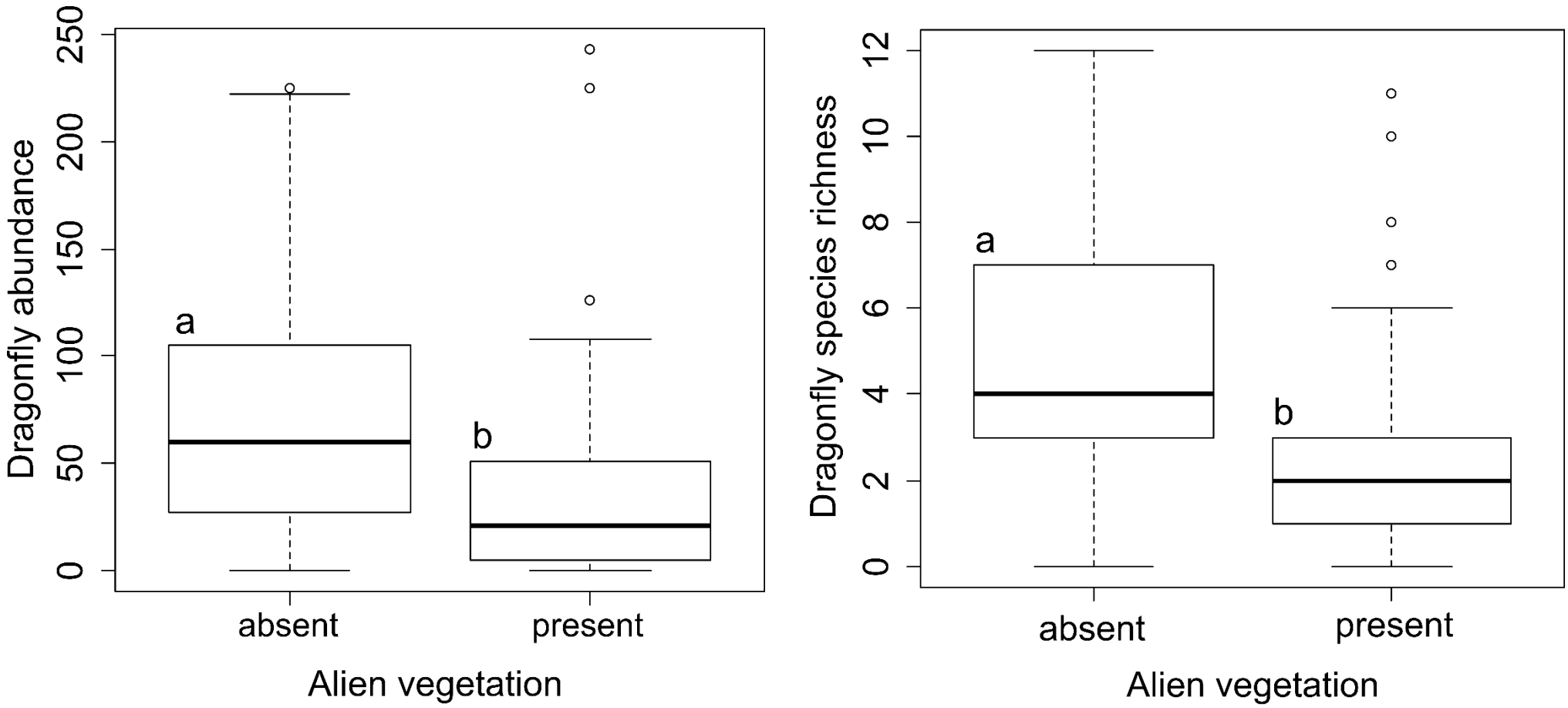


Fig. 5.4 Effect of alien vegetation on dragonfly abundance and species richness and shade cover on species richness.

5.3.4 Effect of environmental variables on dragonfly assemblage composition

All seven variables were retained by the forward selection procedure for dragonfly composition in the CCA (Fig. 5.5). Dragonfly composition and the explanatory variables were significantly related ($P < 0.01$) for the first axis eigenvalue (pseudo-F = 4.60), with 11.7 % of the variance explained by the selected variables and 4.02 % by axis 1. Of the selected variables, alien plant species (EXOT) was the most important variable driving dragonfly assemblages, explaining 2.7% of the variation. Vegetation cover (VEC) and medium height grass (MGRA) were the next most important variables, together explaining 3.9 % of the variation in the dragonfly assemblage.

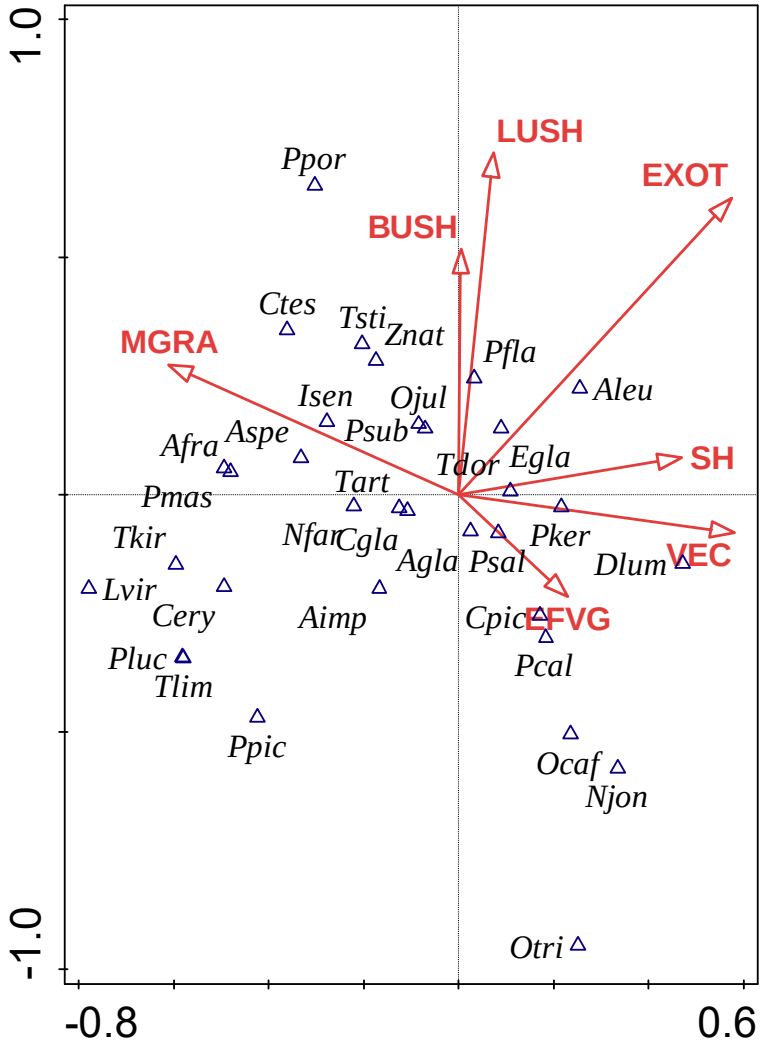


Fig. 5.5 Canonical correspondence analysis ordination showing environmental variables that significantly influenced dragonfly assemblage composition, based on a forward selection procedure. VEC = vegetation cover, SH = shade, EXOT = alien vegetation, LUSH = marginal forest, BUSH = herbs, sedges and broad-bladed grasses, EFVG = emergent vegetation, MGRA = medium grass height.

5.3.5 Unique and shared dragonfly species

There were no species unique to the stream. Four species were only observed during the years after the pond had first been created. Five unique species were recorded at the restored pond. Surprisingly, since pond restoration in 2018 the pond supported an additional six species that were never recorded there previously (Table 5.3). Six species were shared between the stream and original pond that were not recorded in at the restored pond and included: *Lestes plagiatus, Notiothemis jonesi, Orthetrum caffrum, O. trinacria, Palpopleura portia* and *Paragomphus cognatus*. Three species, *Diplacodes luminans, Phyllomacromia picta* and *Pseudagrion massaicum* were recorded in the original and restored pond states, but not from the stream. Only *Ceratogomphus pictus* was recorded at the stream and restored pond but absent from the original pond (Appendix 5.D for species list).

Table 5.3 Table of the species unique between the original pond (1993 and 2001) and the restored pond (2018).

Original pond	**Restored pond**
Agriocnemis falcifera/pinheyi	*Palpopleura lucia*
Orthetrum abbotti	*Lestes virgatus*
Urothemis edwardsi	*Aeshna subpupillata*
Pseudagrion hageni	*Tramea limbata*
	Trithemis kirbyi

5.4 Discussion

Here, I have shown that in an urban landscape, dragonfly diversity is positively influenced by manipulating a stream to include a pond. The initial creation of the pond allowed for an additional seven species to inhabit the waterbody. However, when incorrectly managed and left so that the pond silted up and vegetation overgrew, dragonfly diversity decreased. In such a situation, human intervention in an attempt to restore the artificial pond ecosystem proved to be beneficial for dragonfly diversity. The response of dragonfly assemblages to pond restoration efforts was very rapid, which almost completely recovered even before the project had been completed, adding credence to their use for general studies on aquatic restoration

ecology. Although five species recorded at the initial pond were not present at the restored pond, six other species that had not been recorded before now, inhabited the restored pond. The CCA by Suh and Samways (2005) for the open pond system in 2001 found similar variables to be important for driving the dragonfly assemblage.

The pond state supported the highest dragonfly abundance and species richness. This is in agreement with Suh and Samways (2005), who found that species richness benefitted from pond construction, provided that a variety of biotopes were maintained. For the well-managed pond, habitat heterogeneity was provided in terms of both water and vegetation characteristics. The habitat heterogeneity hypothesis predicts higher species diversity in more structurally complex habitats, due to the provision of more niches (Bazzaz, 1975; Tews et al., 2004). When the stream was impounded to create a pond, and when restored, habitat conditions became suitable to accommodate both lentic and lotic dragonfly species. Species such as *P. massaicum* favours still water conditions and was thus only recorded when the pond was present (Samways & Simaika, 2016). Management of the system also included the establishment of various flow regimes, like riffles, three weirs and a waterfall (The Witness, 2018). Here, and in the prior study by Suh and Samways (2005), dragonfly composition was strongly influenced by vegetation cover, which co-varied with flow regime. Flow rate is a well-known driver of dragonfly assemblages, with different species having specific biotope preferences (Stewart & Samways, 1998).

Area size and habitat heterogeneity are usually positively correlated, and their influences are difficult to distinguish (MacArthur & Wilson, 1967; Rosenzweig, 1995; Báldi, 2007). The area available for occupancy was physically increased by pond creation, and so the system could support a greater number of dragonflies, both adults and presumably their aquatic larvae. This is predominantly important for the territorial males, as pond construction would have increased the number of territories available to be colonized (Parr, 1983). The increased area would also provide habitats for additional aquatic invertebrates. Given that dragonflies are insectivores, a greater abundance of prey would sustain a higher number of individuals (Pritchard, 1965; Duong et al., 2017). Additionally, pond management maximised plant diversity, which would increase invertebrate diversity, particularly herbivores (Siemann et al., 1998).

When a pond, heterogeneity in terms of vegetation characteristics included both structural and compositional aspects. The influence of vegetation cover may be a direct representation of

vegetation succession. For the same pond, Suh and Samways (2005) recognized that around the pond the different stages of vegetation succession were responsible for the high dragonfly diversity. However, when left unmanaged, succession can have the opposite effect, as seen here, where increased vegetation cover decreased dragonfly abundance and species richness and also influenced community composition. The region falls within the grassland biome and over time, succession of tall grasses and establishment of alien plant species (which were positively correlated) may have outcompeted other indigenous vegetation, so reducing heterogeneity with regards to plant diversity and structure (Kinvig & Samways, 2000).

Alien plant species, especially trees, are detrimental to dragonfly diversity mostly due to habitat shading (Samways & Taylor, 2004; Kietzka et al., 2015). Shade reduces species richness and was at its highest during the years when the system had reverted back to a stream. Dragonflies are ectothermic, so many species avoid shaded habitats and are sensitive to the different microclimates produced by vegetation (McGeoch & Samways, 1991; Samways et al., 2005; Samways, 2006; Valentine et al., 2007; Remsburg et al.,2008). When the open pond was well-managed in 2001, alien plant species did not have an influence on dragonflies but shade still played an important role in shaping their assemblage composition. Here, alien plant species decreased dragonfly species richness an influenced assemblage composition. *Pontederia* spp. were the most common invasive plants, and although not trees, they grow in dense clumps which obstruct access to water, outcompete indigenous riparian species, and can disrupt flow (Invasive Species South Africa, 2018).

Shortly after the pond had been restored, dragonfly abundance recovered to the same level as when the pond was first created and well-managed. As adults, dragonflies are highly mobile, observant and base habitat selection mostly on visual cues, which is why they quickly recognised the improved waterbody state and colonised it (Olberg et al., 2005; Remsburg et al., 2008). Furthermore, species richness had almost completely recovered too, even though there were differences in which species were present or absent between the originally created pond and the restored pond. Some species, such as *U. edwardsi*, are geographically marginal for the area, and thus irregular in their appearances. Alternatively, species differences between the original pond and the restored pond may have been due to interspecific interference competition, which is common between adult dragonflies (Moore, 1964; Samways et al., 1996). Species, such *P. portia*, were present when the pond was first created and when it was a stream, but absent from the restored pond. On the other hand, *P. lucia* was only present at the restored

pond. This is likely be due to niche overlap between the two species, as they both have very similar microhabitat preferences (Simaika & Samways, 2016). This phenomenon is more common between congeneric species, as a result of past conflicts such as those observed between *P. kersteni* and *P. hageni*, as well as between *O. julia* and *O. caffrum* (Meskin, 1989; Samways et al., 1996).

5.5 Conclusions

The short- and long-term benefits associated with conservation ponds and succession within urban landscapes have been documented in Europe and South Africa. They have shown that the negative influences on aquatic biota, particularly dragonflies, brought on by urbanisation can be improved through pond construction along an existing stream. Urban landscapes should thus build ponds along streams and manage them in a way that maximises habitat heterogeneity in all its forms in order to accommodate the requirements of many different species. Recommendations for pond management highlight the importance of avoiding adverse ecological succession. This would require maintaining vegetation structure and composition, as well as preventing alien vegetation from invading. To further benefit dragonfly diversity, management of the system should aim to maximize the amount of open, sunlit areas, and provide a range of flow regimes. As dragonflies are an umbrella taxon for other co-occurring species, we would expect that their diversity would also benefit from such management applications in urban landscapes.

5.6 References

Auerswald, K., & Geist, J. (2018). Extent and causes of siltation in a headwater stream bed: catchment soil erosion is less important than internal stream processes. Land Degradation and Development, 29(3), 737-748.

Báldi, A. (2008). Habitat heterogeneity overrides the species–area relationship. Journal of Biogeography, 35(4), 675-681.

Balzan, M. V. (2012). Associations of dragonflies (Odonata) to habitat variables within the Maltese Islands: a spatio-temporal approach. Journal of Insect Science, 12, 87.

Bates, D., Maechler, M., Bolker, B., & Walker, S. (2014). lme4: Linear mixed-effects models using Eigen and S4. R package version, 1(7), 1-23.

Bazzaz, F. A. (1975). Plant species diversity in old-field successional ecosystems in southern Illinois. Ecology, 56, 485-488.

Bolker, B. M., Brooks, M. E., Clark, C. J., Geange, S. W., Poulsen, J. R., Stevens, M. H. H., & White, J. S. S. (2009). Generalized linear mixed models: a practical guide for ecology and evolution. Trends in Ecology and Evolution, 24(3), 127-135.

Bried, J., Herman, B., & Ervin, G. (2007). Umbrella potential of plants and dragonflies for wetland conservation: a quantitative case study using the umbrella index. Journal of Applied Ecology, 44, 833-842.

Briers, R. A., & Biggs, J. (2003). Indicator taxa for the conservation of pond invertebrate diversity. Aquatic Conservation: Marine and Freshwater Ecosystems, 13(4), 323-330.

Canty, A., & Ripley, B. (2012). boot: Bootstrap R (S-Plus) functions. R package version, 1(7).

Chovanec, A. (1994). Man-made wetlands in urban recreational areas - a habitat for endangered species?. Landscape and Urban Planning, 29, 43-54.

Chovanec, A., & Raab, R. (1997). Dragonflies (Insect, Odonata) and the ecological status of newly created wetlands - examples for long-term bio-indication programmes. Limnologica, 27, 381-392.

Corbet, P. S. (1999). Dragonflies: behavior and ecology of Odonata. Harley Books, Colchester, South Africa.

Córdoba-Aguilar, A. (2008). Dragonflies and damselflies: model organisms for ecological and evolutionary research. Oxford University Press, Oxford, UK.

Duong, T. M., Gomez, A. B., & Sherratt, T. N. (2017). Response of adult dragonflies to artificial prey of different size and colour. PloS one, 12(6), e0179483.

Frainer, A., Polvi, L. E., Jansson, R., & McKie, B. G. (2018). Enhanced ecosystem functioning following stream restoration: the roles of habitat heterogeneity and invertebrate species traits. Journal of Applied Ecology, 55(1), 377-385.

Gittleman, J. L., & Kot, M. (1990). Adaptation: statistics and a null model for estimating phylogenetic effects. Systematic Zoology, 39(3), 227-241.

Harabiš, F., & Dolný, A. (2012). Human altered ecosystems: Suitable habitats as well as ecological traps for dragonflies (Odonata): The matter of scale. Journal of Insect Conservation, 16(1), 121-130.

Hassall, C., & Anderson, S. (2015). Stormwater ponds can contain comparable biodiversity to unmanaged wetlands in urban areas. Hydrobiologia, 745, 137-149.

Hill, M. J., Ryves, D. B., White, J. C., & Wood, P. J. (2016). Macroinvertebrate diversity in urban and rural ponds: Implications for freshwater biodiversity conservation. Biological Conservation, 201, 50–59.

Hothorn, T., Bretz, F., & Westfall, P. (2008). Simultaneous inference in general parametric models. Biometrical Journal, 50(3), 346–363.

Invasive Species South Africa. (2018). Protecting Biodiversity from Invasion - Pickerel weed | Pontederia cordata. (n.d), Retrieved from http://www.invasives.org.za/legislation/item/311-pickerel-weed-pontederia-cordata.

Johnson, N., Revenga, C., & Echeverria, J. (2001). Managing water for people and nature. Science, 292(5519), 1071-1072.

Kietzka, G. J., Pryke, J. S., & Samways, M. J. (2015). Landscape ecological networks are successful in supporting a diverse dragonfly assemblage. Insect Conservation and Diversity, 8(3), 229-237.

Kietzka, G. J., Pryke, J. S., & Samways, M. J. (2017). Aerial adult dragonflies are highly sensitive to in-water conditions across an ancient landscape. Diversity and Distributions, 23(1), 14-26.

Kietzka, G. J., Pryke, J. S., & Samways, M. J. (2018). Comparative effects of urban and agricultural land transformation on Odonata assemblages in a biodiversity hotspot. Basic and Applied Ecology (in press).

Kinvig, R. G., & Samways, M. J. (2000). Conserving dragonflies (Odonata) along streams running through commercial forestry. Odonatologica, 29(3), 195-208.

Legendre, P., & Legendre, L. (1998). Numerical ecology: developing in environmental modelling 20. Elsevier, Amsterdam, Netherlands.

Lepš, J., & Šmilauer, P. (2003). Multivariate Analysis of Ecological Data using CANOCO. Cambridge University Press, Cambridge, UK.

MacArthur, R.H., & Wilson, E.O. (1967). The theory of island biogeography. Princeton University Press, Princeton, USA.

Maynou, X., Martin, R., & Aranda, D. (2017). The role of small secondary biotopes in a highly fragmented landscape as habitat and connectivity providers for dragonflies (Insecta: Odonata). Journal of Insect Conservation, 21(3), 517-530.

Mazerolle, M. J., & Mazerolle, M. M. J. (2017). Package 'AICcmodavg'. R package.

McGeoch, M., & Samways, M. (1991). Dragonflies and the thermal landscape: implications for their conservation (Anisoptera). Odonatologica, 20(3), 303-320.

Meskin, I. (1989). Aspects of territorial behaviour in three species of Pseudagrion, Selys (Zygoptera: Coenagrionidae). Odonatologica, 18, 253-261.

Moore, N. W. (1964). Intra- and interspecific competition among dragonflies (Odonata): an account of observations and field experiments in population density in Dorset. Journal of Animal Ecology, 33, 49-71.

Moore, N. W. (1991). The development of dragonfly communities and the consequences of territorial behaviour: a 27-year study of small ponds at Woodwalton Fen, Cambridgeshire, United Kingdom. Odonatologica, 20, 203-231.

Moore, N. W. (2001). Changes in the dragonfly communities at the twenty ponds at Woodwalton Fen, Cambridgeshire, United Kingdom, since the study of 1962-1988. Odonatologica, 30, 289-298.

Oertli, B. (2008). The use of dragonflies in the assessment and monitoring of aquatic habitats. In dragonflies and damselflies: model organisms for ecological and evolutionary research. Oxford University Press (pp. 79-95), Oxford, United Kingdom.

Ogle, D. H. (2018). FSA: fisheries stock analysis. R package version 0.8. 20.9000.

Oksanen, J., Kindt, R., Legendre, P., O'Hara, B., Simpson, G. L., Solymos, P., Stevens, M. H., & Wagner, H. (2008). The vegan package version 1.15-0.

Olberg, R. M., Worthington, A. H., Fox, J. L., Bessette, C. E., & Loosemore, M. P. (2005). Prey size selection and distance estimation in foraging adult dragonflies. Journal of Comparative Physiology, 191(9), 791-797.

Osborn, R., & Samways, M. J. (1996). Determinants of adult assemblage patterns at new ponds in South Africa. Odonatologica, 25, 49-58.

Palmer, M. W. (1993). Putting things in even better order - the advantages of canonical correspondence-analysis. Ecology, 74, 2215-2230.

Parr, M. J. (1983). An analysis of territoriality in libellulid dragonflies (Anisoptera: Libellulidae). Odonalologica (Vol. 12). Retrieved from http://natuurtijdschriften.nl/download?type=document;docid=591603

Primack, R., Kobori, H., & Mori, S. (2000). Dragonfly pond restoration promotes conservation awareness in Japan. Conservation Biology, 14(5), 1553-1554.

Pritchard, G. (1965). Prey capture by dragonfly larvae (Odonata; Anisoptera). Canadian Journal of Zoology, 43(2), 271-289.

Pryke, J. S., & Samways, M. J. (2008). Conservation of invertebrate biodiversity on a mountain in a global biodiversity hotspot, Cape Floral Region. Biodiversity and Conservation, 17(12), 3027.

R Core Team. (2016). R: A language and environment for statistical computing. Vienna: R Foundation for Statistical Computing. Available: http://www.R-project.org.

Remsburg, A. J., Olson, A. C., & Samways, M. J. (2008). Shade alone reduces adult dragonfly (Odonata: Libellulidae) abundance. Journal of Insect Behavior, 21(6), 460-468.

Rosenzweig, M.L. (1995). Species diversity in space and time. Cambridge University Press, Cambridge, UK.

Salemi, L. F., Lins, S. R. M., Ravagnani, E. D. C., Magioli, M., Martinez, M. G., Guerra, F., Vidas, N. B., Fransozi, A., de Barros Ferraz, S. F., & Martinelli, L. A. (2016). Past and present land use influences on tropical riparian zones: an isotopic assessment with implications for riparian forest width determination. Biota Neotropica, 16(2).

Samways, M. J. (2006). Threat levels to odonate assemblages from invasive alien tree canopies. Dragonflies and Forest Ecosystems, 209-244.

Samways, M. J., Caldwell, P. M., & Osborn, R. (1996). Spatial patterns of dragonflies (Odonata) as indicators for design of a conservation pond. Odonatologica, 25(2), 157-166.

Samways, M. J., & Simaika, J. P. (2016). Handbook of freshwater health: the dragonfly Biotic Index for South Africa. South African National Biodiversity Institute, Pretoria, South Africa.

Samways, M. J., & Taylor, S. (2004). Impacts of invasive alien plants on Red-Listed South African dragonflies (Odonata): working for water. South African Journal of Science, 100(1-2), 78-80.

Samways, M. J., Taylor, S., & Tarboton, W. (2005). Extinction reprieve following alien removal. Conservation Biology, 19(4), 1329-1330.

Schmidt, E. (1985). Habitat inventorization, characterisation and bio-indication by a representative spectrum of Odonata species (RSO). Odonatologica, 14, 127-133.

Siemann, E., Tilman, D., Haarstad, J., & Ritchie, M. (1998). Experimental tests of the dependence of arthropod diversity on plant diversity. The American Naturalist, 152(5), 738-750.

Simaika, J. P., & Samways, M. J. (2009a). An easy-to-use index of ecological integrity for prioritizing freshwater sites and for assessing habitat quality. Biodiversity and Conservation, 18, 1171-1185.

Simaika, J. P., & Samways, M. J. (2009b). Reserve selection using Red Listed taxa in three global biodiversity hotspots: dragonflies in South Africa. Biological Conservation, 142(3), 638-651.

Simaika, J. P., & Samways, M. J. (2011). Comparative assessment of indices of freshwater habitat conditions using different invertebrate taxon sets. Ecological Indicators, 11(2), 370-378.

Stewart, D. A. B., & Samways, M. J. (1998). Conserving dragonfly (Odonata) assemblages relative to river dynamics in a major African savanna game reserve. Conservation Biology 12, 683-692.

Steytler, N. S., & Samways, M. J. (1995). Biotope selection by adult male dragonflies (Odonata) at an artificial lake created for insect conservation in South Africa. Biological Conservation, 72(3), 381-386.

Suh, A. N., & Samways, M. J. (2001). Development of a dragonfly awareness trail in an African botanical garden. Biological Conservation, 100(3), 345-353.

Suh, A. N., & Samways, M. J. (2005). Significance of temporal changes when designing a reservoir for conservation of dragonfly diversity. Biodiversity and Conservation, 14(1), 165-178.

ter Braak, C. J. F. (1990). Interpreting canonical correlation analysis through biplots of structure correlations and weights. Psychometrika, 55, 519-531.

ter Braak, C. J. F., & Šmilauer, P. (2012). CANOCO Reference Manual and User's Guide: Software for Ordination (version 5.0). Microcomputer Power, Ithaca, USA.

Tews, J., Brose, U., Grimm, V., Tielbörger, K., Wichmann, M. C., Schwager, M., & Jeltsch, F. (2004). Animal species diversity driven by habitat heterogeneity/diversity: the importance of keystone structures. Journal of Biogeography, 31(1), 79-92.

The Witness. (2018). What is new at Kingfisher Lake. Available: https://www.pressreader.com/south-africa/the-witness/20180611/281483572082954.

Toms, J. D., & Lesperance, M. L. (2003). Piecewise regression: a tool for identifying ecological thresholds. Ecology, 84(8), 2034-2041.

Valentine, L. E., Roberts, B., & Schwarzkopf, L. (2007). Mechanisms driving avoidance of non-native plants by lizards. Journal of Applied Ecology, 44(1), 228-237.

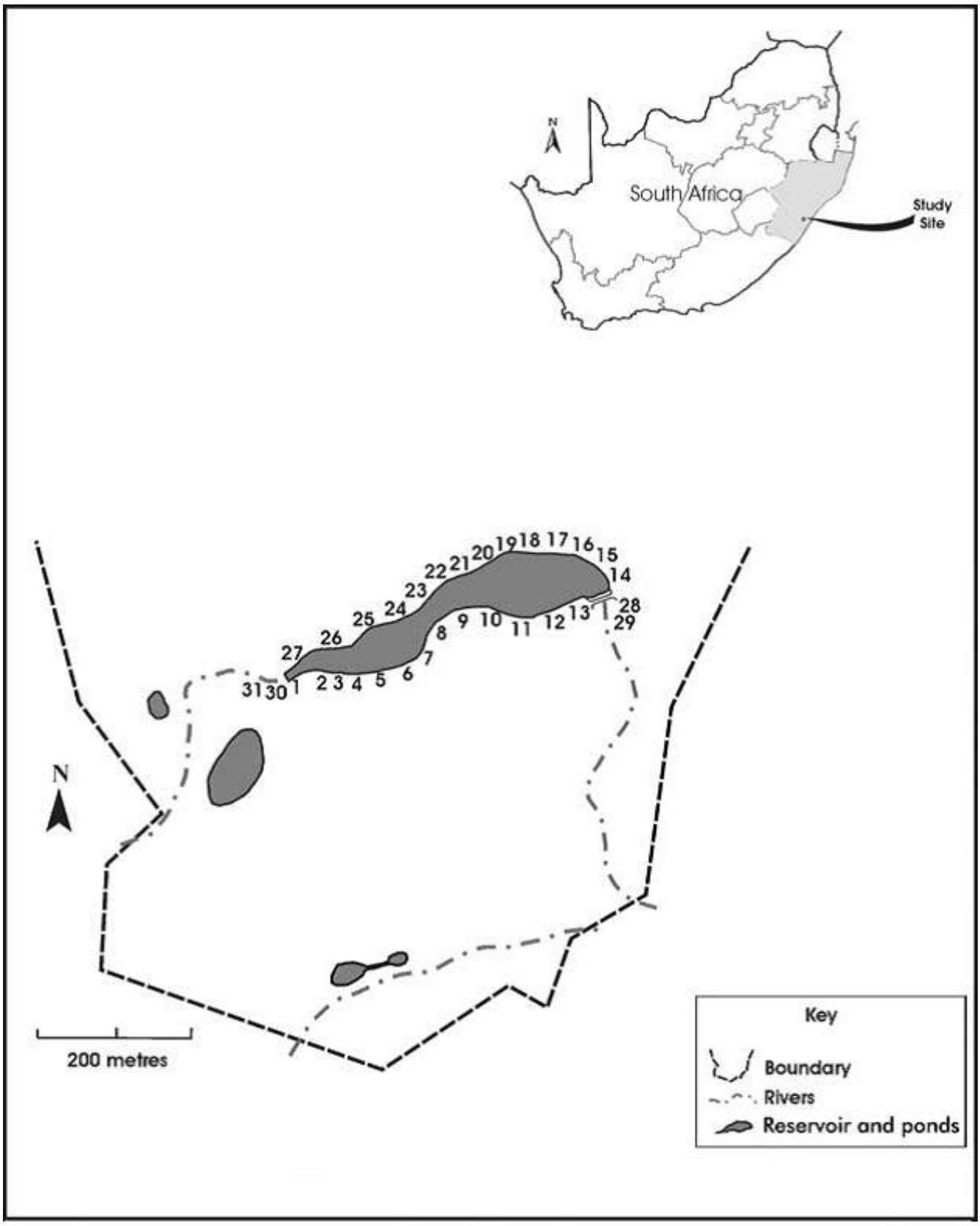
Willis, C. K., & Samways, M. J. (2011). Water Dancers of South Africa's National Botanical Gardens. SANBI, Pretoria, South Africa.

Willis, C., & Samways, M. J. (2013). Dragonfly & Damselfly Trail Guide: KwaZulu-Natal National Botanical Garden. SANBI, Pretoria, South Africa.

Wood, P. J., & Armitage, P. D. (1997). Biological effects of fine sediment in the lotic environment. Environmental Management, 21(2), 203-217.

5.7 Appendices

Appendix 5.A Map of Pietermaritzburg Botanical Gardens site showing the pond circumference divided into 27 sampling units (SUs) with SUs 28 and 29 at the outflow stream/ waterfall and SUs 30 and 31 positioned upstream. Map taken from Suh and Samways (2005).

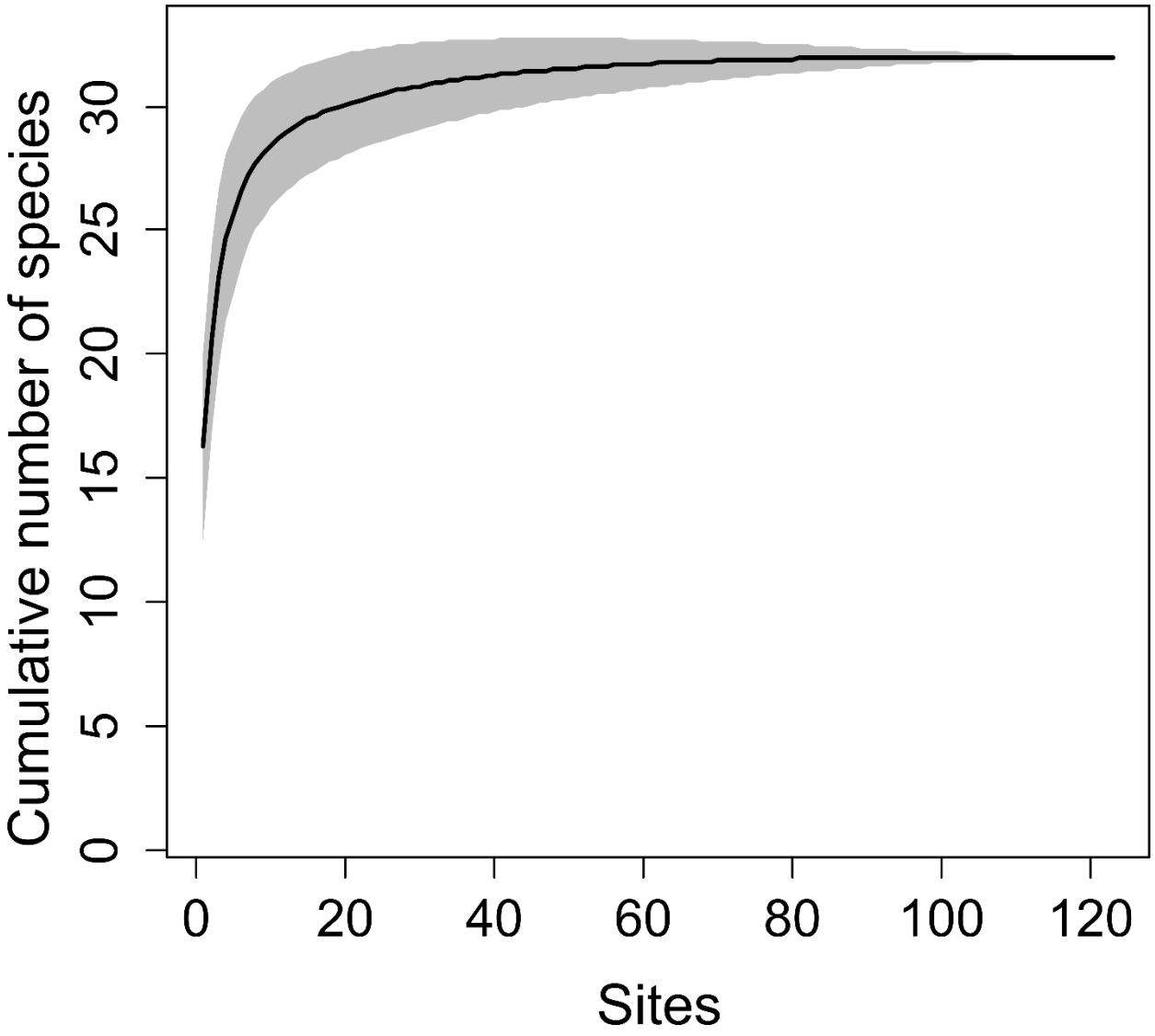


Appendix 5.B Photographs of the unmanaged pond that reverted to a stream A) and shortly after restoration B).

A)

B)

Appendix 5.C Rarefaction curve for the dragonflies sampled in this study.



Appendix 5.D List of species present for the original pond, the stream and the restored pond. Presence of a species marked with an X.

Species scientific name	Original pond	Stream	Restored pond
Africallagma fractum (Ris 1921)	X	X	X
Africallagma glaucum (Burmeister 1839)	X	X	X
Agriocnemis falcifera (pinhey 1959)	X	-	-
Allocnemis leucosticta (Selys 1863)	X	X	X
Anax imperator (Rambur 1842)	X	X	X
Anax speratus (Hagen 1867)	X	X	X
Ceratogomphus pictus (Selys 1854)	-	X	X
Ceriagrion glabrum (Burmeister 1839)	X	X	X
Chlorolestes tessellatus (Burmeister 1839)	X	X	X
Crocothemis erythraea (Brullé 1832)	X	X	X
Diplacodes luminans (Karsh 1893)	X	-	X
Elattoneura glauca (Selys 1862)	X	X	X
Ischnura senegalensis (Rambur 1842)	X	X	X
Lestes plagiatus (Burmeister 1839)	X	X	-
Lestes virgatus (Burmeister 1839)	-	-	X
Nesciothemis farinosa (Förster 1898)	X	X	X
Notiothemis jonesi (Ris 1919)	X	X	-
Orthetrum abbotti (Calvert 1892)	X	-	-
Orthetrum caffrum (Burmeister 1839)	X	X	-
Orthetrum Julia falsum (Longfield 1955)	X	X	X
Orthetrum trinacria (Selys 1841)	X	X	-
Palpopleura lucia (Drury 1773)	-	-	X
Palpopleura portia (Drury 1773)	X	X	-
Pantala flavescens (Fabricius 1798)	X	X	X
Paragomphus cognatus (Rambur 1842)	X	X	-
Phyllomacromia picta (Hagen in Selys 1871)	X	-	X
Pinheyschna subpupillata (McLachlan 1896)	-	-	X
Platycypha caligata (Selys 1853)	X	X	X
Pseudagrion hageni (Pinhey 1966)	X	-	-
Pseudagrion kersteni (Gersaecker 1869)	X	X	X
Pseudagrion massaicum (Sjöstedt 1909)	X	-	X
Pseudagrion salisburyense (Ris 1921)	X	X	X
Tramea limbata (Desjardins 1832)	-	-	X
Trithemis arteriosa (Burmeister 1839)	X	X	X
Trithemis dorsalis/furva (Rambur 1842/Karsch 1899)	X	X	X
Trithemis kirbyi (Selys 1891)	-	-	X
Trithemis stictica (Burmeister 1839)	X	X	X
Urothemis edwardsi (Selys 1849)	X	-	-
Zygonyx natalensis (Martin 1900)	X	X	X

Chapter 6: General conclusions

Anthropogenic impacts cause severe degradation of freshwater ecosystems (Vörösmarty et al., 2010). This necessitates urgent conservation action that requires rapid, reliable assessment methods. Bioindication, or the use of certain species that reflect the biotic or abiotic condition of an ecosystem, the influence of environmental change on communities, or the diversity of other taxa (McGeoch, 1998; Gerhardt, 2002), has gained popularity as a cost-effective approach for effectual assessments. It constitutes an important sub-discipline within the broader field of conservation biology, with the final objective to aid ecosystem management based on bioindicator knowledge (McGeogh, 1998). For freshwater ecosystems, adult dragonflies are increasing in popularity as bioindicators (Schindler et al., 2003). They inhabit a wide variety of biotopes, are taxonomically well known, variably sensitive to habitat changes and are easy to identify and sample (Samways & Steytler, 1996; Chovanec & Waringer, 2001). However, much still needs to be done to realise their full potential as an indicator taxon. This is important as management decisions made using bioindicators need to be taken in light of their abilities and limitations. Therefore, this study focussed on exploring new ways of expanding the use of adult dragonflies as bioindicators.

Dragonflies are best known for their potential as environmental and ecological indicators. Although suggested as biodiversity indicators (Simaika & Samways, 2012), little research has been done to assess this application. Biodiversity indicators are taxa that represent the species richness of other co-occurring taxa, but it is impossible to define a single indicator or group to represent all of biodiversity (Fleishman, Murphy & Brussard, 2000; Vandewalle et al., 2010). The value of biodiversity indication is perceptible when applied to biodiversity conservation (Duelli & Obrist, 2003). I have shown for the first time that adult dragonflies are successful biodiversity indicators for the Ephemeroptera, Plecoptera and Trichoptera (EPT) of four Greater Cape Floristic Region (GCFR) rivers, in the Western Cape Province of South Africa. However, using the full complement of dragonfly taxa in a monitoring program can be costly and time consuming, which contradicts the purpose of bioindicators. A subset of the biodiversity indicators were therefore quantitatively selected. Using the umbrella index (Fleishman, Murphy & Brussard, 2000), I was able to identify seven dragonflies whose preservation would result in the conservation of most EPT taxa. Furthermore, they were also good surrogates for the conservation of overall dragonflies. As a result, managers would only

need to monitor this select group of dragonfly species to increase the successful conservation of dragonfly and EPT taxa in these rivers.

Previously, the umbrella index had only been tested on birds and butterflies (Betrus, Fleishman & Blair, 2005), and on dragonflies and wetland plants in the USA (Bried et al., 2007). To thoroughly test the limits of this index, I applied it to four rivers with varying degrees of anthropogenic disturbance. These are located in a biodiversity hotspot renowned for its heterogeneous river ecosystems, with a large number of endemic and rare aquatic invertebrates (Dallas & Day, 2007). Furthermore, the taxa used are highly sensitive to disturbances and were at different stages of their life cycles (adult, terrestrial dragonflies and the aquatic larvae of EPT). The umbrella index proved its flexibility even when faced with the aforementioned challenges (Fleishman et al., 2000). Moreover, its ability to perform across a spatial scale greater than a single river is a particularly important criterion for continued use in future conservation planning.

The index also identified a group of EPT taxa that were comparatively just as effective as surrogates for the overall dragonflies as the identified dragonfly umbrella species (Chapter 2). The spatial associations between EPT and dragonfly groups suggested that conservation managers could choose either one for sufficient biodiversity planning (Chapter 2). Although the focus of this study was on the use of dragonflies as indicators, knowledge of the interchangeability of the two groups is useful. For example, when conditions are not suitable for dragonfly sampling, managers could utilize EPT umbrellas to convey protection to their own group and for the overall dragonflies here.

Adult dragonflies are only present for a short period during the summer months (van Schalkwyk et al., 2014; Samways & Simaika, 2016; Tarboton & Tarboton, 2019). Like other insects, they are ectothermic and most adults are heliophiles (Paulson, 2019). Due to their dependence on air temperature, they are active in the warmer hours of the day and when weather conditions are fine and windless (Samways & Steytler 1996; Simaika & Samways, 2010). This limits the use of adult dragonflies as bioindicators. There was therefore a need to evaluate the suitability of dragonfly larvae as indicators, as these can be sampled under any weather conditions and at different times of the year (Raebel et al., 2010). The research reported in my work indicated that data on surveys of adults and larvae are not always interchangeable (Chapter 3). A major outcome of this result was that the importance of scale and the type of

question investigated. Many questions related to their use as biondicators are based on rough estimates of assemblages, such as abundance or species richness, which are often conducted at larger spatial scales. The interchangeability of the two life stages is then possible. For example, as an environmental indicator, to investigate the influence of a pollutant along a river, a researcher could compare dragonfly species richness above the pollutant input source to below it. Alternatively, the application of dragonflies in the Dragonfly Biotic Index (DBI) could also be used (Simaika & Samways, 2009; Samways & Simaika, 2016). However, this is not always the case, as with taxonomic surveys of dragonflies or monitoring needed at the point scale. In such instances, or for community-related questions, surveying larvae, adults and exuviae is recommend. This pattern was driven by differences in the habitat requirements of the two life stages.

Adult dragonflies have successfully indicated the negative impacts associated with anthropogenic development (Šigutová et al., 2019). Many studies recommend demarcating protected areas free from any human disturbances to conserve biodiversity (Hermoso et al., 2018; Kietzka et al., 2018; Loury et al., 2018). However, in a world where agricultural and urban development is inevitable, it is becoming increasingly difficult to do so. Additionally, conservationists must aim to maintain biodiversity within transformed landscapes (Minor et al., 2018; Briggs et al., 2019). Therefore, various methods have been applied to conserve freshwater biodiversity in transformed landscapes. Dragonflies have proven themselves in determining the success of some of these methods, and in the identification of the crucial components to consider for present and future management (Samways, 1989; Samways & Steytler, 1996; Kinvig & Samways, 2000; Sayer et al., 2012; Goertzen & Suhling, 2013; Kietzka et al., 2015). In the current work, I used dragonflies to indicate the success of biodiversity conservation-management practices in transformed landscapes, one in forestry plantations (Chapter 4) and one in an urban setting (Chapter 5), in the KwaZulu-Natal Province of South Africa.

Plantation forestry is one of the dominant agricultural activities in KwaZulu-Natal. These exotic, monoculture plantations have detrimental effects on river ecosystems and their biodiversity (Jewitt et al., 2015). Previous studies identified water pollution and shading from trees as the primary contributors to declines in dragonfly diversity and habitat quality (Kinvig & Samways, 2000). Ecological networks (ENs), that include rivers, were established by forestry companies as a means to reduce the negative impacts associated with plantations

(Samways et al., 2010). Here, adult dragonflies validated the success of these ENs, since their diversity did not differ between EN sites and nearby protected areas (PAs) (Chapter 4). ENs also maintained good water quality, as Dragonfly Biotic Index (DBI) scores did not differ between ENs and PAs. For plantations in KwaZulu-Natal, the EN approach proved to be highly effective for the maintenance of dragonfly diversity and habitat quality in lotic systems (Chapter 4). Furthermore, dragonflies identified in-water variables, elevation gradients and corridor width as the most important variables to include in successful EN management and future design. These ENs have also shown to be effective for terrestrial invertebrates, including ants, grasshoppers, spiders and scarab beetles (Bazelet & Samways, 2011; Pryke & Samways, 2012; Pryke et al., 2013). Furthermore, they benefit some specialist bird species, small mammals like the rare oribi and large mammals such as the African elephant and white rhinoceros (Lipsey & Hockey, 2010; Samways & Pryke, 2016).

Green spaces integrated into cities aim to alleviate the damaging impacts of urbanisation on biodiversity (Aronson et al., 2017). Terrestrial species, such a birds, beetles, spiders and butterflies, benefit from green spaces (Dearborn & Kark, 2010; Goddard et al., 2010). These areas can include artificial aquatic features, such as ponds, to benefit freshwater biodiversity, as seen for frogs (Goddard et al., 2010; Hill et al., 2016; 2017). Artificial ponds can also provide important ecosystem services such as improving water quality, nutrient cycling or storm water control (Mallin et al., 2002; Hassall & Anderson, 2015; Holtmann et al., 2018). In lieu of this, an insect and aquatic plant conservation pond was constructed along a stream in the National Botanical Gardens in Pietermaritzburg, KwaZulu-Natal (Suh & Samways, 2001; 2005). I have shown that adult dragonflies can be successfully utilised as indicators of conservation success in this urban environment as it significantly increased dragonfly species richness and abundance, a major objective when creating it in the first place (Suh & Samways, 2005; Chapter 5). Similarly, pond creation in Wales increased the diversity of macrophytes and aquatic invertebrates (Gee et al., 1997).

Due to lack of proper management, over time the aforementioned pond silted up, became overgrown with vegetation and reverted into a narrow stream. It has been shown that dragonflies can be used as indicators of restoration progress, such as following the removal of alien tree species along a river (Magoba & Samways, 2010). Dragonfly diversity (abundance and species richness) at this pond followed the decline in habitat quality during the siltation process. In 2018, the pond was restored to include a variety of microhabitat conditions, similar

to the layout of the original pond. After only a few months, dragonfly diversity had recovered and resembled that of the original pond providing strong evidence for the use of dragonflies as indicators of restoration progress. This was despite the transformed urban environment that was dominated by generalist species. Furthermore, the importance of correctly managing conservation ponds in order to maintain their value became apparent.

Concerning dragonflies and freshwater biodiversity in transformed landscapes, two themes were evident throughout both the altered environments. Firstly, the importance of creating connectivity to alleviate the effects of fragmentation caused by transformed landscapes and the importance of habitat heterogeneity. For the plantations, these were the ENs natural grassland corridors (containing rivers), which connected to each other and to larger PAs. For the botanical garden surrounded by urban sprawl, it was the construction of the pond itself along a narrow stream. Ideally, these structures function as habitats and provide adequate resources and optimal breeding conditions for some species. However, for species that cannot survive in rivers in EN corridors or urban ponds, these structures can act as stepping-stones to enable dispersion through the landscape to more favourable habitats and provide temporary resources in the transition (Simaika et al., 2016). Therefore, future studies should include assessment of the diversity of the aquatic dragonfly larvae and exuviae to determine whether dragonflies use these altered habitats for breeding or as stepping-stones. Either way, the importance of these habitats for the movement of dragonflies should not be underestimated given evidence based on metopopulation dynamics science (Rosenberg et al., 1997; Keymer et al., 2000; Harabiš & Dolny, 2012).

Dragonflies also conveyed the importance of creating heterogeneity in conservation structures in transformed landscapes. In the ENs, this was reflected by the positive correlation between corridor width and dragonfly species richness and abundance. Wider corridors typically harbour higher habitat heterogeneity and would meet the requirements of a greater range of species (Haddad et al., 2003; Samways & Pryke, 2016; van Schalkwyk et al., 2017). Similarly, pond construction also increased habitat heterogeneity. The transformation of a stream to include a pond provided habitats for both lentic and lotic species. Furthermore, the pond included a variety of microhabitats in terms of vegetation-related characteristics and flow dynamics. Applicable to both cases is also the species-area relationship (Montaña et al., 2015). Larger areas naturally contain more resources, such as food, which would be sufficient for an abundance of dragonflies. Furthermore, dragonflies were proven as functional bioindicators for

rivers in corridors of plantations, as well as in an artificial conservation pond in a city. Their ability to perform under two very different circumstances (lotic waters in plantation settings and a lentic waterbody in an urban city) adds to their value as highly flexible indicators.

In conclusion, dragonflies proved their worth as model organisms for conservation research and should be endorsed in conservation management worldwide. Monitoring of dragonflies revealed the importance of protected areas for conserving biodiversity but more importantly, that biodiversity conservation is possible in transformed landscapes. Therefore, I recommend ENs be considered in other agricultural practices and that artificial ponds be implemented into more cities. Dragonflies showed that for the aforementioned as well as for other biodiversity conservation methods, management should maximise habitat heterogeneity and connectivity in order to accommodate many different species and allow for movement across transformed landscapes. To determine the success of other conservation strategies and restoration projects, I endorse the use of dragonflies as bioindicators (Samways & Taylor, 2004; Simaika & Samways, 2009). Particularly in South Africa, more research is required on the interchangeability of the life stages. This includes defining the appropriate sampling methods for each stage under varying conditions. For species-specific, small spatial scale research, it became evident that larvae, adults and exuviae should all be sampled. This has been proposed by the guidelines for the application of the DBI. However, this may result in much longer times needed for sampling and the need for expert knowledge, both that may offset the need for extensive surveying at larger scales and for specific questions. Of the three indicator categories, biodiversity indicators are the least understood as it is impossible to find one single indicator of biodiversity. The conflicting choices for values and measures make it difficult to reach a consensus on the use and choice of biodiversity indicators. Future research should investigate the potential of dragonflies as biodiversity indicators of other taxa including those not necessarily bound to aquatic systems. I recommend applying the umbrella index for quantitative selection and that its usefulness be further tested.

6.1 References

Aronson, M. F., Lepczyk, C. A., Evans, K. L., Goddard, M. A., Lerman, S. B., MacIvor, J. S., Nilon, C. H., & Vargo, T. (2017). Biodiversity in the city: key challenges for urban green space management. Frontiers in Ecology and the Environment, 15(4), 189–196.

Bazelet, C. S., & Samways, M. J. (2011). Grasshopper assemblage response to conservation ecological networks in a timber plantation matrix. Agriculture, Ecosystems and Environment, 144(1), 124–129.

Betrus, C. J., Fleishman, E., & Blair, R. B. (2005). Cross-taxonomic potential and spatial transferability of an umbrella species index. Journal of Environmental Management, 74(1), 79–87.

Bried, J. T., Herman, B. D., & Ervin, G. N. (2007). Umbrella potential of plants and dragonflies for wetland conservation: A quantitative case study using the umbrella index. Journal of Applied Ecology, 44(4), 833–842.

Briggs, A. J., Pryke, J. S., Samways, M. J., & Conlong, D. E. (2019). Complementarity among dragonflies across a pondscape in a rural landscape mosaic. Insect Conservation and Diversity, 12(3), 241–250.

Chovanec, A., & Waringer, J. (2001). Ecological integrity of river-floodplain systems - assessment by dragonfly surveys (Insecta: Odonata). Regulated Rivers: Research and Management, 17(4–5), 493–507.

Dallas, H. F., & Day, J. A. (2007). Natural variation in macroinvertebrate assemblages and the development of a biological banding system for interpreting bioassessment data - A preliminary evaluation using data from upland sites in the south-western Cape, South Africa. Hydrobiologia, 575(1), 231–244.

Dearborn, D. C., & Kark, S. (2010). Motivations for conserving urban biodiversity. Conservation Biology, 24(2), 432 – 440.

Duelli, P., & Obrist, M. K. (2003). Biodiversity indicators: The choice of values and measures. Agriculture, Ecosystems and Environment, 98(1–3), 87–98.

Fleishman, E., Murphy, D., & Brussard, P. F. (2000). A new method for selection of umbrella species for conservation planning. Ecological Applications, 10(2), 569–579.

Gee, J. H. R., Smith, B. D., Lee, K. M., & Griffiths, S. W. (1997). The ecological basis of freshwater pond management for biodiversity. Aquatic Conservation: Marine and Freshwater Ecosystems, 7(2), 91–104.

Gerhardt, A. (2002). Bioindicator species and their use in biomonitoring. Environmental Monitoring, 1, 77 – 123.

Goddard, M. A., Dougill, A. J., & Benton, T. G. (2010). Scaling up from gardens: biodiversity conservation in urban environments. Trends in Ecology and Evolution, 25(2), 90–98.

Goertzen, D., & Suhling, F. (2013). Promoting dragonfly diversity in cities: Major determinants and implications for urban pond design. Journal of Insect Conservation, 17(2), 399–409.

Haddad, N. M., Spira, T., Levey, D. J., Sargent, S., Danielson, B. J., Bowne, D. R., & Cunningham, A. (2003). Corridor use by diverse taxa. Ecology, 84(3), 609–615.

Harabiš, F., & Dolný, A. (2012). Human altered ecosystems: Suitable habitats as well as ecological traps for dragonflies (Odonata): The matter of scale. Journal of Insect Conservation, 16(1), 121–130.

Hassall, C., & Anderson, S. (2015). Stormwater ponds can contain comparable biodiversity to unmanaged wetlands in urban areas. Hydrobiologia, 745, 137 – 149.

Hermoso, V., Thieme, M., Abell, R., Linke, S., & Turak, E. (2018). Defining and enhancing freshwater protected areas. In Freshwater Ecosystems in Protected Areas: Conservation and Management (pp. 54 – 69). Routledge, Oxfordshire, England.

Hill, M. J., Biggs, J., Thornhill, I., Briers, R. A., Gledhill, D. G., White, J. C., Wood, P. J., & Hassall, C. (2017). Urban ponds as an aquatic biodiversity resource in modified landscapes. Global Change Biology, 23(3), 986–999.

Hill, M. J., Ryves, D. B., White, J. C., & Wood, P. J. (2016). Macroinvertebrate diversity in urban and rural ponds: Implications for freshwater biodiversity conservation. Biological Conservation, 201, 50–59.

Holtmann, L., Juchem, M., Brüggeshemke, J., Möhlmeyer, A., & Fartmann, T. (2018). Stormwater ponds promote dragonfly (Odonata) species richness and density in urban areas. Ecological Engineering, 118, 1–11.

Jewitt, D., Goodman, P. S., Erasmus, B. F. N., O'Connor, T. G., & Witkowski, E. T. F. (2015). Systematic land-cover change in KwaZulu-Natal, South Africa: Implications for biodiversity. South African Journal of Science, 111(9–10), 01–09.

Keymer, J. E., Marquet, P. A., Velasco-Hernández, J. X., & Levin, S. A. (2000). Extinction thresholds and metapopulation persistence in dynamic landscapes. The American Naturalist, 156(5), 478 – 494.

Kietzka, G. J., Pryke, J. S., & Samways, M. J. (2015). Landscape ecological networks are successful in supporting a diverse dragonfly assemblage. Insect Conservation and Diversity, 8(3), 229–237.

Kietzka, G. J., Pryke, J. S., & Samways, M. J. (2018). Comparative effects of urban and agricultural land transformation on Odonata assemblages in a biodiversity hotspot. Basic and Applied Ecology, 33, 89–98.

Kinvig, R. G., & Samways, M. J. (2000). Conserving dragonflies (Odonata) along streams running through commercial forestry. Odonatologica, 29(3), 195 – 208.

Lipsey, M. K., & Hockey, P. A. R. (2010). Do ecological networks in South African commercial forests benefit grassland birds? A case study of a pine plantation in KwaZulu-Natal. Agriculture, Ecosystems and Environment, 137(1–2), 133–142.

Loury, E. K., Ainsley, S. M., Bower, S. D., Chuenpagdee, R., Farrell, T., Guthrie, A.G., Heng, S., Lunn, Z., Mamun, A. A., Oyanedel, R., & Rocliffe, S. (2018). Salty stories, fresh spaces: Lessons for aquatic protected areas from marine and freshwater experiences. Aquatic Conservation: Marine and Freshwater Ecosystems, 28(2), 485 – 500.

Magoba, R. N., & Samways, M. J. (2010). Recovery of benthic macroinvertebrate and adult dragonfly assemblages in response to large scale removal of riparian invasive alien trees. Journal of Insect Conservation, 14(6), 627–636.

Mallin, M. A., Ensign, S. H., Wheeler, T. L., & Mayes, D. B. (2002). Pollutant removal efficacy of three wet detention ponds. Journal of Environmental Quality, 31(2), 654 – 660.

McGeogh, M. A. (1998). The selection, testing and application of terrestrial insects as bioindicators. Biological Reviews, 73(2), 181–201.

Minor, E. S., Anderson, E. C., Belaire, J. A., Garfinkel, M., & Smith, A. D. (2018). Urban green infrastructures and ecological networks for urban biodiversity conservation. In urban biodiversity (Vol. 186, No. 199, pp. 186 – 199). Routledge, London, England.

Montaña, C. G., Layman, C. A., & Winemiller, K. O. (2015). Species-area relationship within benthic habitat patches of a tropical floodplain river: An experimental test. Austral Ecology, 40(3), 331–336.

Paulson, D. (2019). Dragonflies and damselflies: A natural history. Princeton University Press, New Jersey, USA.

Pryke, J. S., Roets, F., & Samways, M. J. (2013). Importance of habitat heterogeneity in remnant patches for conserving dung beetles. Biodiversity and Conservation, 22(12), 2857–2873.

Pryke, J. S., & Samways, M. J. (2012). Ecological networks act as extensions of protected areas for arthropod biodiversity conservation. Journal of Applied Ecology, 49(3), 591–600.

Raebel, E. M., Merckx, T., Riordan, P., Macdonald, D. W., & Thompson, D. J. (2010). The dragonfly delusion: why it is essential to sample exuviae to avoid biased surveys. Journal of Insect Conservation, 14(5), 523 – 533.

Rosenberg, D. K., Noon, B. R., & Meslow, E. C. (1997). Biological corridors: form, function, and efficacy. BioScience, 47(10), 677 – 687.

Samways, M. J. (1989). Farm dams as nature reserves for dragonflies (Odonata) at various altitudes in the Natal Drakensberg Mountains, South Africa. Biological Conservation, 48(3), 181–187.

Samways, M. J., Bazelet, C. S., & Pryke, J. S. (2010). Provision of ecosystem services by large scale corridors and ecological networks. Biodiversity and Conservation, 19(10), 2949–2962.

Samways, M. J., & Pryke, J. S. (2016). Large-scale ecological networks do work in an ecologically complex biodiversity hotspot. Ambio, 45(2), 161–172.

Samways, M. J., & Simaika, J. P. (2016). Manual of freshwater assessment: Dragonfly biotic index. South African Biodiversity Institute, Pretoria, South Africa.

Samways, M. J., & Steytler, N. S. (1996). Dragonfly (Odonata) distribution patterns in urban and forest landscapes, and recommendations for riparian management. Biological Conservation, 78(3), 279 – 288.

Samways, M. J., & Taylor, S. (2004). Impacts of invasive alien plants on Red-Listed South African dragonflies (Odonata). South African Journal of Science, 100(1 – 2), 78 – 80.

Sayer, C., Andrews, K., Shilland, E., Edmonds, N., Edmonds-Brown, R., Patmore, I., Emson, D., & Axmacher, J. (2012). The role of pond management for biodiversity conservation in an agricultural landscape. Aquatic Conservation: Marine and Freshwater Ecosystems, 22(5), 626–638.

Schindler, M., Fesl, C., & Chovanec, A. (2003). Dragonfly associations (Insecta: Odonata) in relation to habitat variables: a multivariate approach. Hydrobiologia, 497(1 – 3), 169 – 180.

Šigutová, H., Šipoš, J., & Dolný, A. (2019). A novel approach involving the use of Odonata as indicators of tropical forest degradation: When family matters. Ecological Indicators, 104, 229–236.

Simaika, J. P., & Samways, M. J. (2009). An easy-to-use index of ecological integrity for prioritizing freshwater sites and for assessing habitat quality. Biodiversity and Conservation, 18(5), 1171–1185.

Simaika, J. P., & Samways, M. J. (2010). Comparative assessment of indices of freshwater habitat conditions using different invertebrate taxon sets. Ecological Indicators, 11, 370–378.

Simaika, J. P., & Samways, M. J. (2012). Using dragonflies to monitor and prioritize lotic systems: a South African perspective. Organisms Diversity and Evolution, 12(3), 251 – 259.

Simaika, J. P., Samways, M. J., & Frenzel, P. P. (2016). Artificial ponds increase local dragonfly diversity in a global biodiversity hotspot. Biodiversity and conservation, 25(10), 1921 – 1935.

Suh, A. N., & Samways, M. J. (2001). Development of a dragonfly awareness trail in an African botanical garden. Biological Conservation, 100(3), 345–353.

Suh, A. N., & Samways, M. J. (2005). Significance of temporal changes when designing a reservoir for conservation of dragonfly diversity. Biodiversity and Conservation, 14(1), 165–178.

Tarboton, W., & Tarboton, M. (2019). Field guide to dragonflies and damselflies. Penguin Random House South Africa, Cape Town, South Africa.

van Schalkwyk, J., Samways, M. J., & Pryke, J. S. (2014). Winter survival by dragonfly adults in the Cape Floristic Region. International Journal of Odonatology, 17(1), 17–30.

van Schalkwyk, J., Pryke, J. S., & Samways, M. J. (2017). Wide corridors with much environmental heterogeneity best conserve high dung beetle and ant diversity. Biodiversity and Conservation, 26(5), 1243–1256.

Vandewalle, M., De Bello, F., Berg, M. P., Bolger, T., Doledec, S., Dubs, F., Feld, C. K., Harrington, R., Harrison, P. A., Lavorel, S., & Da Silva, P. M. (2010). Functional traits as indicators of biodiversity response to land use changes across ecosystems and organisms. Biodiversity and Conservation, 19(10), 2921–2947.

Vörösmarty, C. J., McIntyre, P. B., Gessner, M. O., Dudgeon, D., Prusevich, A., Green, P., Glidden, S., Bunn, S. E., Sullivan, C. A., Liermann, C. R., & Davies, P. M. (2010). Global threats to human water security and river biodiversity. Nature, 467(7315), 555–561.